

# MYCOLOGIA

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## NEW TROPICAL FUNGI

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(WITH PLATES 18-21)

### *Shropshiria* Stevens, gen. nov.

Sori on the periphery of sclerotoid stromata, open, bordered by sterile peridia; conidiophores long, simple, bearing the conidia laterally. Conidia 1-celled, globose, smoky.

Named in honor of Mr. J. B. Shropshire in recognition of his helpful interest in the biologic sciences in the Panama Canal Zone.

### *Shropshiria Chusqueae* Stevens, sp. nov.

Sporiferous stromata caulicolous, globose, or subglobose to irregularly oblong, brown to gray or black, 3-8 mm. in diameter, roughened over the entire surface by the projecting spore columns. Sporogenous cups about 150-180  $\mu$  wide at the open mouth, about 230  $\mu$  deep, and dark bordered on sides and bottom. Sterile space between cups 150-230  $\mu$ . The rind between the sori in surface view, microscopically, is light brown in color and made up of cells of quite uniform size, about 3-4  $\mu$  in diameter and having a pseudo-parenchymoid appearance. Conidiophores arising from the brown base of the cup, hyaline, parallel and straight, slender, simple, septate, about 2  $\mu$  thick, 250  $\mu$  long. The spores are borne laterally on the distal ends of the conidiophores. Conidia smoky, globose, dark in mass, 3  $\mu$  in diameter.

On *Chusquea simpliciflora*.

Panama. Brazos Brook reservoir, 9-22-1924. No. 697 (type); Culebra, 10-2-1924. No. 939; Las Cruces trail, 9-28-1924. No. 876.

The sclerotoid stromata always develop laterally on the stem of the bamboo and remain lateral, *i.e.*, they do not encircle them.

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In young specimens they are smooth and brown, but as they become sporogenous they are thickly beset with the sori, which to the naked eye are gray in their centers, and thus the whole stroma assumes a grayish appearance. Each sorus appears as a slightly raised papilla distinctly cup-shaped (PLATE 18, FIG. 2; PLATE 21, FIG. 1) with a thick, dark border.

The sclerotium is of almost flinty consistency, such that it is almost impossible to cut it. By boiling in dilute potash it is however softened to a cheesy consistency and may be readily cut by razor or imbedded in paraffine and cut on the microtome. Boiling in dilute lactic acid is also helpful. In microscopic view of the sclerotium in section the greater part of the interior of the mass is seen to be composed of intricately interwoven, coarse, irregular, hyaline mycelium. The periphery only is differentiated into a cortical or rind region. This is quite dark brown, about  $18\ \mu$  thick and is subtended by an inner cortex, less dark, about  $50\ \mu$  thick which merges indefinitely into the central plexus. The second or inner cortex is composed of fine hyphae, about  $2\ \mu$  thick, *i.e.*, about half as thick as the coarse hyaline mycelium composing the body of the structure.

The sori, I use this term merely with the significance of spore beds, are of genuine cup-shape, resembling in general aspect the cup of *Aecidium*. They are sunken into the periphery of the stroma, and their sides, peridia, project very slightly above the level of the surrounding stroma. On sides and bottom the sori are bordered by a dark layer, of much the same texture and color as the rind of the stroma, this layer merging somewhat indefinitely into the body of the stroma.

While the border of the sorus is composed of crooked interwoven hyphae the central region of the cup is filled with the slender, long, parallel conidiophores. They are hyaline, or very faintly straw colored, and are divided by many septa into cells which are quite uniformly  $5-6\ \mu$  long. These are not conidiiferous in the basal portion, but are so for a considerable distance at the distal end.

Though very small and thus very difficult of observation it appears that the spores are borne laterally on the conidiophores on short sterigmata, probably more than one (2-3 or 4) arising

at one node of the conidiophores. The conidia are produced in great numbers and rest in heaps on each sorus.

The relationship of this fungus is problematic. Were it to be considered on general principles it appears to me that it must be assigned to the Fungi Imperfecti. There, however, it shows no close relationship with either of the three large orders. It certainly is not pycnidial. I do not think the sorus can properly be regarded as an acervulus though there is some superficial resemblance to one. There remains only the Moniliales where it would have to fall either with the Moniliaceae or the Stilbaceae with neither of which does it agree well. There is no genus in any of the groups mentioned above that presents characters showing even remote relationship to this genus. *Coniosporium*, which is sometimes mentioned as resembling *Graphiola* in its mode of bearing spores, certainly shows no similarity with our fungus.

There is one genus, however, sometimes placed in the Fungi Imperfecti, viz., *Graphiola*, with which it does show certain important characters of agreement though there are many important differences. The points of resemblance are as follows: the manner in which the conidia are borne, pleurogenous, on the conidiophores; the close parallel arrangement of the conidiophores, fertile only on the distal portion; the cup-like receptacle in which the conidiophores are borne; the dark border of the peridium bounding the sorus; parasitic on a monocotyledonous plant.

The points of difference from *Graphiola* are as follows: the well-developed sclerotoid stroma with the sori sunken in its periphery; the lack of a parallel arrangement of the mycelium of the peridium; the light color of the peridium; the absence of long sterile conidiophores among the fertile (not always present in *Graphiola*); the dark color of the conidia.

The differences from *Graphiola* are certainly strongly marked; nevertheless, in the absence of any evident relationship with any other group of fungi, it appears to me that the agreements in character indicated above warrant the placing of this genus in the Graphiolaceae.

The relationship of *Graphiola* has long been in doubt, it having

been variously assigned to the Uredinales, the Ustilaginales, the Pyrenomycetes and the Fungi Imperfecti. An extensive investigation by E. Fisher<sup>1</sup> working under DeBary in 1883 led him to the conclusion that *Graphiola*, while not to be placed in the Ustilaginales, showed nearest relation to this order. *Stylina*, another genus in the Graphiolaceae, as characterized by Sydow,<sup>2</sup> has several sori in a stroma and the sterile hyphal bundles are lacking, thus rendering this fungus somewhat nearer to ours in character than is *Graphiola*. *Farysia*, which shows superficial resemblances to *Graphiola* in the possession of sterile hyphal bundles<sup>3</sup> and in some other detail of structure, appears to present a very different manner of spore formation from that of the Graphiolaceae. Sydow and Butler<sup>4</sup> describing *Graphiola Borassi* refer to the sterile filaments. They cite the spores as being produced on the upper segments of the fertile hyphae as 3 to 8 linear protrusions, usually four. The sterile bundles between the fertile are clearly shown in their figures. It is different from *G. Phoenicis* in that it long remains covered. The mode of spore production in this species appears to differ somewhat from our form in that the separate cells of the conidiophores fall apart.

The form sometimes called *Graphiola macrospora* Penz. & Sacc., also *Melanconium profundus* Penz. & Sacc.,<sup>5</sup> should properly be placed in the genus *Endocalyx* as *E. melanoxanthus* Berk. & Br. Its spores are black and its essential morphology is so different from *Graphiola* that it need not be further considered.

In the new system of von Höhnelt<sup>6</sup> the genus *Graphiola* is placed in the Stromataceen-Angiostromaceae, Pachystromaceen-Sphaeriales-Jacenta-Coriaceae. Independent of our conclusion

<sup>1</sup> Fisher, Ed. Beitrag zur Kenntniss der Gattung *Graphiola*. Bot. Zeit. 41: 745. 1883.

<sup>2</sup> In Fischer, Ed. Zur Kenntnis von *Graphiola* und *Farysia*. Ann. Myc. 18: 188. 1920.

<sup>3</sup> Raciborski, M. Parasitische und epiphytische Pilze Javas. Bull. Acad. Sci. Cracovie 1909: 346-394. 1909.

<sup>4</sup> Sydow, H. & P., & Butler, E. J. Fungi Indiae orientalis. Ann. Myc. 5: 489. 1907.

<sup>5</sup> Petch, T. The genus *Endocalyx*, Berkeley and Broome. Ann. Bot. 22: 389. 1908.

<sup>6</sup> Höhnelt, F. von. System der Fungi imperfecti Fuckel. Mykol. Unters. Berichte 1: 301. 1923.



as to the relationship of our fungus to *Graphiola* it appears to fall in the von Höhnelt system in a position near to *Graphiola*.

If it be tentatively admitted that this fungus belongs to the Graphiolaceae its differences from the previously known forms in that group are so great that little or no suggestion of phylogeny can be apparent until other intermediate forms are found.

**Clypeodiplodina** Stevens, gen. nov.

Pycnidia immersed, early thickened to a clypeus above, astomate, opening by rupture and fragmentation of the upper wall. Conidia oblong, hyaline, 1-septate.

**Clypeodiplodina Baccharidis** Stevens, sp. nov.

Pycnidia roughly circular, 150–520  $\mu$  in diameter, about 150  $\mu$  deep, closed when young, later opening by rupture. Mouth of open pycnidium 90–390  $\mu$  in diameter, internal wall about 10–18  $\mu$  thick, hyaline, external edge up to 55  $\mu$  thick and 90  $\mu$  long, densely black. Hyaline wall sporiferous throughout, black wall sterile. Conidiophores short, 3–4  $\mu$ , from a hyaline pseudoparenchyma. Conidia hyaline, irregular, 25–40  $\times$  7  $\mu$ , 1-septate when mature.

On *Baccharis floribunda* (parasitic on the living leaves).

Ecuador, Guapulo, Nov. 12, 1924. No. 267.

The black pycnidia are usually clustered in irregular groups that may cover a whole leaf or half of it or any portion of it (PLATE 20). The groups are very irregular in outline and arrangement, sometimes occupying all of one side of a leaf, or all of a base, or all of a tip, the other portions being quite free of fungus. The disease appears to be progressive in that the younger pycnidia are at the edges of the pycnidial groups.

In very old groups the pycnidia appear to coalesce so that their individuality is lost and merely a thick black crust remains. In this stage the pycnidia cease to function, no longer bearing conidia, and the black crust appears to be composed of numerous globose aggregations, densely black on the outside and hyaline within, appearing to be sclerotia. It is possible that these later develop asci, but diligent search has failed to reveal such.

The pycnidia develop subepidermally in the mesophyll, but as they grow, compress the overlying cells and eventually replace them

by the clypeus-like thickening of the distal part of the pycnidial wall. Frequently the clypeus attains to  $80\ \mu$  in thickness before rupturing. The conidiophores are short, mere projections from the hymenial cells. Black setae are quite common though not constant on the pycnidia. In some instances they appear to arise from a black superficial mycelium. It is possible, though not probable, that they belong to this fungus.

The relationship of this fungus is somewhat questionable. Observed in its younger stages of development it is a nearly globose, pycnidium-like structure, with a well-defined wall surrounding it, while the upper part of the wall immediately under the host epidermis is darkened and thickened. At this stage it appears clearly to belong to the Sphaeropsidales, though the entire absence of an ostiole is somewhat unusual. In mature form the spore-bearing region gives no hint of its earlier covered condition, but is wide open, cup-shaped, and would appear to belong possibly to the acervuli fungi, though sections show that its admission to that group is precluded by the walls of the sorus. One turns then to the Leptostromataceae or Excipulaceae as possibilities. Both of these groups are poorly characterized and contain at present many fungi that should properly be placed elsewhere. The genera of the Hyalodidymae, in those two families, which are nearest to our fungus are *Leptothyrella* and *Discella* respectively.

Our fungus cannot be *Leptothyrella* which is radiate nor does it appear to be nearly related to *Discella*, most species of which are saprophytes on wood and of structure quite different from ours. Both of these genera also present the flat base which probably should be regarded as truly characteristic of these two orders. In the Sphaerioideae-hyalodidymae our fungus would fall near *Diplodina*, but is widely separated from it by the absence of an ostiole; cup-like character of the mature sorus; and what is perhaps most significant, by the clypeus-like structure of the upper wall of the pycnidium and later the thick, black, peridium-like lip which surrounds the sporogenous region.

The bottom of the pycnidium both in immature and mature forms is never flattened, but is always concave, which character alone seems to me sufficient to prevent placing it in the Lepto-

stromataceae or Excipulaceae and results in placing it in the Sphaerioidaceae. Following the system of von Höhnel<sup>7</sup> our fungus would go to Pycnidiaceen, Pycnidieen, Sphaerioideae-astomae, which in the Hyalodidymae has only two genera, *Pucciniospora* and *Scaphidium*, both quite distinct from ours. In view of these facts it is proposed as a new genus of the Sphaerioideae-hyalodidymae, the most distinctive character being the clypeus on the young pycnidia, which develops to form the large thick lip to the open disc at maturity.

**Chaetothyriopsis** Stevens & Dorman, gen. nov.

Thallus radiate, superficial, no free mycelium; perithecia setose, single, ostiolate, round, brown; spores 1-septate, hyaline; asci clavate, paraphysate, 8-spored.

Similar to *Microthyrium* but setose.

**Chaetothyriopsis panamensis** Stevens & Dorman, sp. nov.

Perithecium 40–70  $\mu$  in diameter, radiate, ostiolate, setae usually 3–4, simple, septate and arising mostly from the dark border of the ostiole, 36–70  $\mu$  long, 4  $\mu$  thick at base, tapering gradually from base to tip, dark at base, pale at tip; asci 21–22  $\times$  7  $\mu$ , clavate, 8-spored; spores hyaline, 1-septate, elliptical, inordinate, 7  $\times$  2  $\mu$ .

On the upper surface of the leaves of *Oncoba laurina*. Panama, Darien, Sept. 10, 1924. No. 411.

This fungus is quite invisible except with the aid of a compound microscope, nor are there any appearances of parasitism. It was discovered merely by accident when searching the leaf for other fungi. The perithecium bears resemblance to that of *Actinopeltis* but our fungus is not parasitic on another fungus, nor do its spores agree at all with those of *Actinopeltis*. The ostiole is very definitely bordered by about three rows of very dark cells.

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<sup>7</sup> Höhnel, F. von. System der Fungi imperfecti Fuckel. Mycol. Unters. Berichte 1: 301. 1923.

## EXPLANATION OF PLATES

## PLATE 18

Fig. 1. *Shropshiria*: somewhat diagrammatic view, showing location of the sori around the periphery of the sclerotium.

Fig. 2. *Shropshiria*: section of a sporogenous cup, showing wall, base, and the parallel conidiophores in the central region.

Fig. 3. *Clypeodiplodina*: drawing, diagrammatic, showing various shapes of pycnidia, and ostioles of various ages.

Fig. 4. *Clypeodiplodina*: an unopened pycnidium showing the distinct surrounding wall, especially thickened at the top.

Fig. 5. *Clypeodiplodina*: section of an old pycnidium showing its cup-like shape and the large development of the black pseudoparenchymatous border.

Fig. 6. *Clypeodiplodina*: detail of pycnidial wall, conidiophores and conidia.

Fig. 7. *Clypeodiplodina*: conidia showing shape and septation.

Fig. 8. *Chaetothyriopsis*: three perithecia showing the radiate structure and the setae.

Fig. 9. *Chaetothyriopsis*: an ascus and spores.

## PLATE 19

*Shropshiria*: photograph showing four stromata about natural size.

## PLATE 20

*Clypeodiplodina*: photograph showing general aspect of the fungus on the leaves.

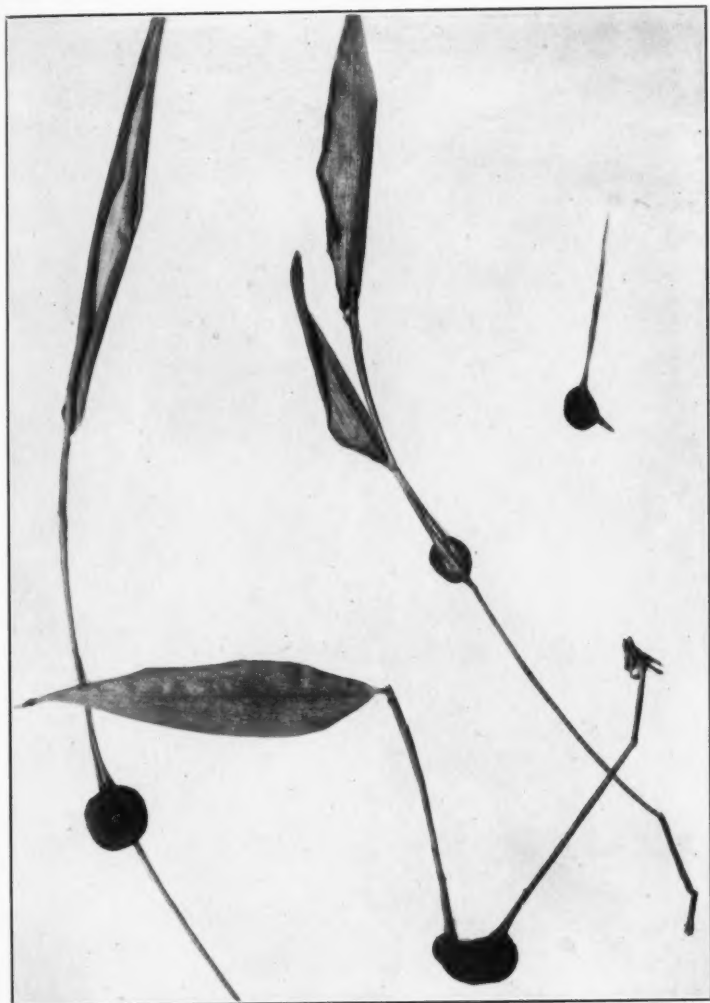
## PLATE 21

Fig. 1. *Shropshiria*: photograph of a single stroma enlarged to show the sori.

Fig. 2. *Clypeodiplodina*: photograph of a leaf cleared by boiling in dilute potash, showing the individual pycnidia.

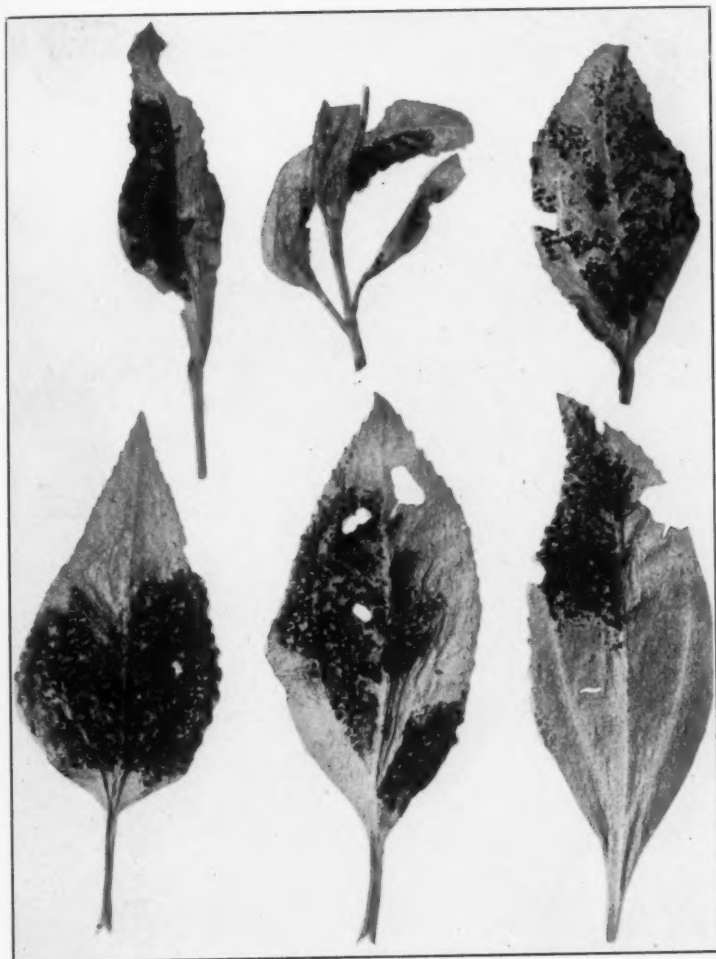
Fig. 3. *Clypeodiplodina*: photograph showing older pycnidia of more complex structure than those of Fig. 2.

Fig. 4. *Clypeodiplodina*: photomicrograph of a pycnidium in median section.

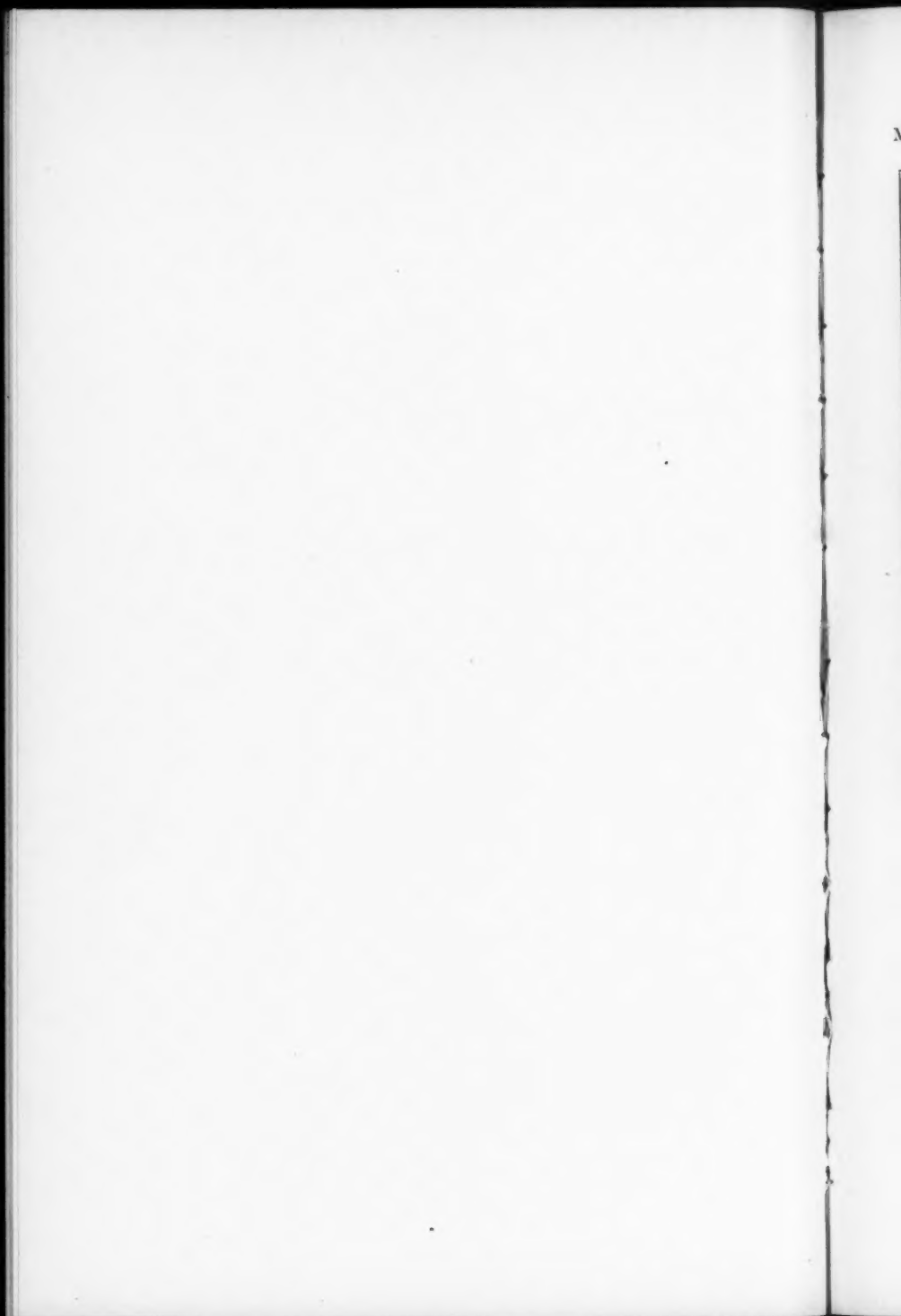


SHROPSHIRIA

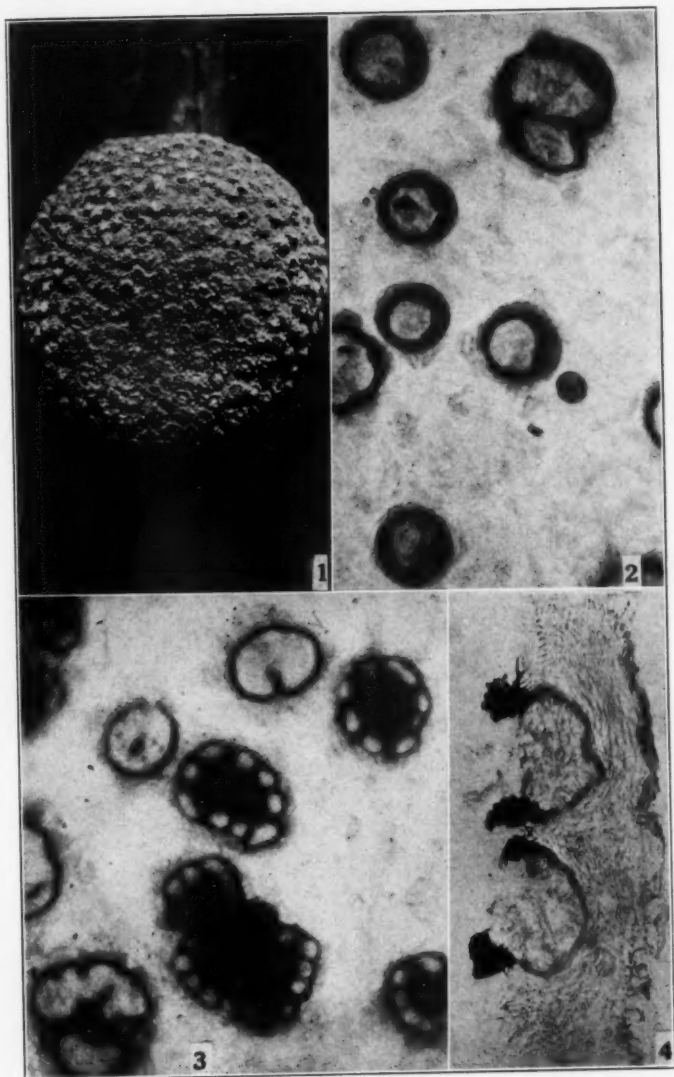




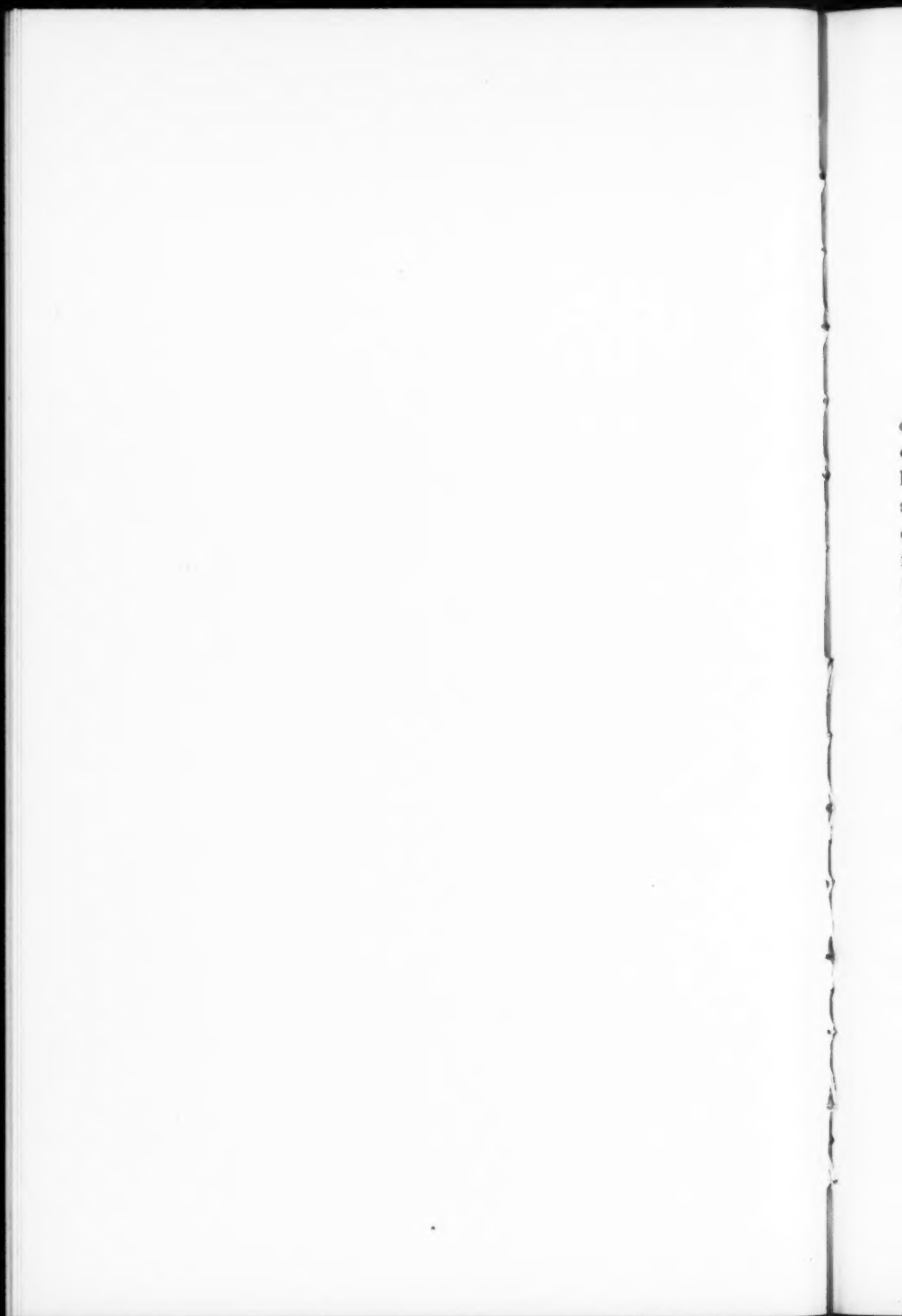
CLYPEODIPLODINA







SHROPSHIRIA AND CLYPEODIPLODINA



## BASIDIA AND SPORES OF THE NIDULARIACEAE

G. W. MARTIN

(WITH PLATES 22 AND 23)

Ever since the classic work of the Tulasnes (11), the position of the Nidulariaceae as Basidiomycetes has been definitely established. Later authors have contributed much to our knowledge of the taxonomy and morphology of the group, but surprisingly little has been added to what the Tulasnes published concerning the basidia. This is partly to be explained by the fact that the time during which spores may be found on the basidia is comparatively short, and the spores are shed and the basidia collapse long before the peridioles are exposed. The Tulasnes note that after their discharge the basidiospores of *Cyathus striatus* develop a thick spore wall, and Sachs (8) observed a similar change in the spores of *Crucibulum vulgare*, but these observations seem not to have been extended to other species. Besides the authors cited, R. E. Fries (4, 5) and Walker (12) have published excellent figures of the basidia of the Nidulariaceae, but these were incidental to morphological and cytological studies, and were made from fixed and stained sections. It seemed desirable, therefore, to make a comparative study of the basidia of as many species as possible. The present paper is based on the examination of three species of *Cyathus*, one of *Crucibulum*, and one of *Nidularia*, of all of which living as well as dried material was available. The dried specimens were soaked in water for at least an hour, then very thin freehand sections were cut and mounted in dilute potassium hydroxide solution, sometimes stained lightly with eosin. Living material was sectioned similarly and either examined untreated or killed in Gram's Iodine solution and stained with eosin. In all five species studied, it was found that the basidia appear when the basidiocarp is less than half its full size. They are at first

clavate or ovate and usually arranged in an hymenial region, lining the inner side of the peridiole wall. They bear their spores and shed them through a brief period, and then completely disappear, leaving the spores immersed in a gelatinous matrix in the center of the peridiole. The spores are not discharged, but are forced from the basidia by the swelling and gelatinization of the tissues lining the peridiole wall, and either preceding or immediately following such separation, the basidia themselves collapse and become gelatinized. After leaving the basidia, the spores undergo a further development. This may involve nothing more than a thickening of the outer spore wall, or it may result in a very great enlargement of the spore itself, continuing after the exposure of the peridiole. It is worthy of note that the spores of all three species of *Cyathus* examined are sessile, while those of *Nidularia* and *Crucibulum* are borne on sterigmata. This would tend to confirm the opinion, previously advanced on other grounds, that *Nidularia* and *Crucibulum* are more closely related to each other than to *Cyathus* and that they are the more primitive genera.

More detailed observations are given under the notes on the individual species.

#### CRUCIBULUM VULGARE Tul.

The basidia are borne in clusters, each with a clamp connection at its base. As the basidium develops, the outer wall, except at the extreme tip, becomes highly gelatinized (PLATE 23, FIGS. 47, 49), and the protoplasm is restricted to a narrow lumen, giving it more of a clavate or stalked appearance than its actual dimensions justify. Four sterigmata are formed and a swelling at the tip of each grows into a spore. In some cases, the gelatinized basidia shrivel without giving rise to spores (PLATE 23, FIG. 48). When the spores have reached their full size, the gelatinous outer wall of the basidium has, as a rule, become indistinguishable. All but one of the basidia observed were four-spored (PLATE 23, FIGS. 50-55). The exception (PLATE 23, FIG. 56) bore two spores, one of which was much larger than the other, and was clearly abnormal. The sterigmata are often of unequal lengths. While the spore is attached to the sterigma,

there is no trace of a hilum. Immediately after its separation there is a slight projection at one end which later disappears. This evidently represents nothing more than the place of attachment. The spores are not discharged, but are forced from the sterigmata and toward the center of the peridiole by the gelatinization of the underlying tissues.

The Tulasnes illustrate a number of basidia of this species, several of them two- or three-spored and some appearing abnormal.

#### NIDULARIA PULVINATA (Schw.) Fries

This is the species called *Nidularia pisiformis* by Lloyd (6) and *Granularia pulvinata* by Miss White (13). The basidia are usually clustered as in *Crucibulum* (PLATE 22, FIG. 3), but they are also borne singly. The gelatinization is apparent at an early stage (PLATE 22, FIGS. 1, 2) and is even more extreme than in the preceding form. While most of the basidia are three- or four-spored (PLATE 22, FIGS. 3, 7-10), a number of two-spored basidia (PLATE 22, FIGS. 5, 6) and a few bearing but one spore (PLATE 22, FIG. 4) were observed. Many of the spores, when freed from the basidia, carry with them the major portion of the sterigma upon which they were borne (PLATE 22, FIG. 11). In *Nidularia pulvinata* the hymenium is more definite than in any of the other species examined.

The Tulasnes (11) illustrate basidia of *Nidularia australis* and *N. Duriacana*, showing them as two- and three-spored and with short sterigmata, and Fries (4, 5) shows basidia of *N. pisiformis* with four, three and two spores on rather long sterigmata.

#### CYATHUS STRIATUS Pers. var. SCHWEINITZII Tul.

The basidia are clavate, with very long stalks so that the basal septum is immersed in a gelatinous mass and difficult to see. I was unable to determine whether or not a clamp connection is present, as in the other forms studied. The basidia are regularly four-spored (PLATE 22, FIGS. 18-20), the spores sessile, rather bluntly elliptical, and at the time of leaving the basidium with a very thin wall (PLATE 22, FIG. 21). Later they develop a thick wall (PLATE 22, FIG. 22) and sometimes a faint, but distinct yellowish tint. This is somewhat accentuated by adding

KOH to the mount. There is usually a rather indefinite, hyaline gelatinous sheath around each spore, difficult to show in a drawing. There is usually evidence of increase in size after separation from the basidia. Spores from mature basidiocarps, while differing somewhat in size and shape, are much less variable than are those of *C. stercoreus*.

CYATHUS OLLA Pers.

The basidia are narrowly clavate or subcylindrical, long stemmed, and, in some cases at least, with a clamp connection at the basal septum. They bear two, three, or four sessile and broadly oval spores (PLATE 22, FIGS. 12-15). Immediately after separation from the basidia, the point of attachment is indicated by a small apiculus, which soon disappears. Many basidia collapse with the spores still attached to them (PLATE 22, FIG. 15). The spores are later separated by the usual process and the basidia eventually disappear. There is no evidence that the spores increase materially in size after leaving the basidia, but the wall may become somewhat thicker.

CYATHUS STERCOREUS (Schw.) De-Toni

The basidia are borne in clusters as in *Crucibulum* and *Nidularia*. They are broadly clavate, the upper part often being nearly globose, with a very short stalk and a prominent clamp connection at the base. The basidia are mostly four-spored. None were seen bearing less than four spores, and many with more, up to eight (PLATE 22, FIGS. 23-28). Abnormal basidia, forked or branching (PLATE 22, FIG. 24), are not uncommon, but none were seen bearing spores. Detachment of the spores is as usual in the group (PLATE 22, FIGS. 29-32). At the time of detachment they are broadly oval, and thin-walled, with a marked apiculus indicating the point of detachment. They are fairly uniform in size at this time, although even on the same basidium there may be considerable variation in this respect. They are commonly 8-10  $\mu$  in length and somewhat less in width at the time the basidia collapse and they are forced into the gelatinous interior of the peridiole. Here each spore is surrounded by a group of specialized hyphae. These hyphae

apparently arise from the inner surface of the peridiole between the basidia, although their origin is not easy to trace since all but the tips soon become empty of protoplasm. The tips are filled with dense cytoplasm and are often richly branched. They are closely appressed to the surface of the spore and obviously serve as nurse hyphae (PLATE 23, FIGS. 39-46). Under these circumstances, the spores undergo a remarkable development, increasing greatly in size, forming a thick outer wall and becoming subglobose or by pressure irregular in shape. In one instance, the outermost peridiole was removed from a young basidiocarp the epiphragm of which was just splitting. The spores were subglobose, thin-walled and with a marked apiculus, and were from 11 to 14  $\mu$  broad (PLATE 23, FIG. 37). They were surrounded by nurse hyphae, not shown in the illustration. The basidiocarp, attached to its substratum, was placed in a moist chamber, and ten days later the second peridiole was removed and the spores examined. They had enlarged to nearly three times their former diameter, all trace of the apiculus had disappeared, and the wall had become thickened (PLATE 23, FIG. 38). The nurse cells had virtually disappeared. The increase in bulk represented approximately 2600 per cent.

The very great variability in the size of the spores of this species has often been noted. Variability is illustrated by the outlines of groups of spores from four different basidiocarps collected at different times and places (PLATE 23, FIGS. 33-36). It seems obvious that the limit of size which the spores might attain in any given case would be greatly influenced by environmental conditions, particularly the amount of moisture in the substratum and in the air, after the spores are separated from the basidia. The only previous instances on record among the Basidiomycetes in which spores increase in size after separation from the basidia seem to be in *Scleroderma* and related genera. This phenomenon was first reported by the Tulasnes (10). They describe the spores as separating from the basidia while the wall is still smooth, and becoming surrounded by a mass of adventitious hyphae, under the influence of which they increase in size and develop the familiar rough walls of the genus. These results were confirmed by Bonorden (2) but denied by Sorokine

(9). Rabinowitsch (7) confirmed the findings of the Tulasnes and Bonorden in general, but describes the hyphae surrounding the spores as nearly empty and often collapsed, and does not believe that they play any significant part in the increase in spore size, accepting instead Wiesner's theory that the spore wall contains living protoplasm and grows by a process of intussusception. Beck (1) found a situation in *Phlyctospora* similar to that reported for *Scleroderma*, but in this case the enveloping hyphae remain attached to the spore, forming a surrounding hull at maturity. In all of these instances, the nature of the hyphae described seems to be similar to that of those here called the nurse hyphae of *Cyathus stercoreus*, but in none of them do spores increase so greatly in size after separation from the basidia.

#### SPORE DISSEMINATION IN CRUCIBULUM

No account, based on observation, has ever been given of the dissemination of spores by the bird's-nest fungi. Brefeld (3), upon theoretical considerations, decided that the spores of *Crucibulum vulgare* must be disseminated by animals eating the peridioles and voiding them, or the spores, in their feces. This view is adopted by deBary. Early in November, 1926, I was collecting Gasteromycetes in a sandy field near Iowa City. This had been planted to corn, but had not been ploughed for several years and was reverting to prairie. Half buried in the ground were the bases of the old cornstalks, nearly every one of which supported a number of basidiocarps of *Crucibulum vulgare*. Above and around every cluster of basidiocarps, on twigs, dead leaves, and similar fragments, were numerous peridioles, obviously forced out of the cups by rain drops, and glued firmly to whatever they had hit. Debris of this sort, readily transported by wind and water, and often deposited in heaps of similar material, would afford highly efficient dissemination of the fungus. Some of the fruiting bodies were brought into the laboratory and soaked for a while. When removed from the water, the peridioles could easily be spattered out of the cups by dropping water into them from a pipette a couple of feet above them. In *Cyathus stercoreus*, as in *Crucibulum*, the funiculus tends to



disappear as the peridioles age, and in this species, as in *Nidularia*, it is possible that rain may play the same rôle. In *Cyathus striatus*, on the other hand, where the funiculus is persistent and the peridioles are well down in the narrow part of the cup, this method of dissemination might be unavailable.

#### SUMMARY

The basidia and spores of five species belonging to the Nidulariaceae, including three species of *Cyathus*, one of *Nidularia* and one of *Crucibulum*, have been studied. The spores of *Cyathus* are sessile; those of *Nidularia* and *Crucibulum* are borne on well-developed sterigmata. The spores of all species are separated from the basidia by the collapse and gelatinization of the latter accompanied by the gelatinization of the tissues lining the walls of the peridiole. After separation, the spores remain immersed in a gelatinous matrix in the interior of the peridiole, where they undergo further development, which may consist in nothing more than a thickening of the spore wall, or may involve a very considerable enlargement of the spore itself, amounting in the case of *Cyathus stercoreus* to 2600 per cent.

A method of dissemination of the peridioles of *Crucibulum* is described.

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## EXPLANATION OF PLATES

All drawings made with camera lucida. Figures 1-11 with Zeiss objective X and ocular K 3, at a magnification of 1530. Figures 12-57 with Zeiss objective D and ocular 4 at a magnification of 720. All reduced approximately one third in reproduction.

## EXPLANATION OF PLATES

## PLATE 22

## NIDULARIA PULVINATA

1. Young basidium; 2. Young basidium with sterigmata developed; 3. Cluster of basidia; 4-10. Basidia with one, two, three, and four spores; 11. Two spores immediately after separation from basidia; sterigmata still attached.

## CYATHUS OLLA

12-14. Mature basidia with two, three, and four spores; 15. Basidium collapsing with spores still attached; 16. Spores immediately after separation from basidia, showing conspicuous apiculus; 17. Older spores; apiculus nearly or quite gone.

## CYATHUS STRIATUS

18-20. Mature basidia, each bearing four spores; 21. Spores immediately after separation from basidia; 22. Mature spores.

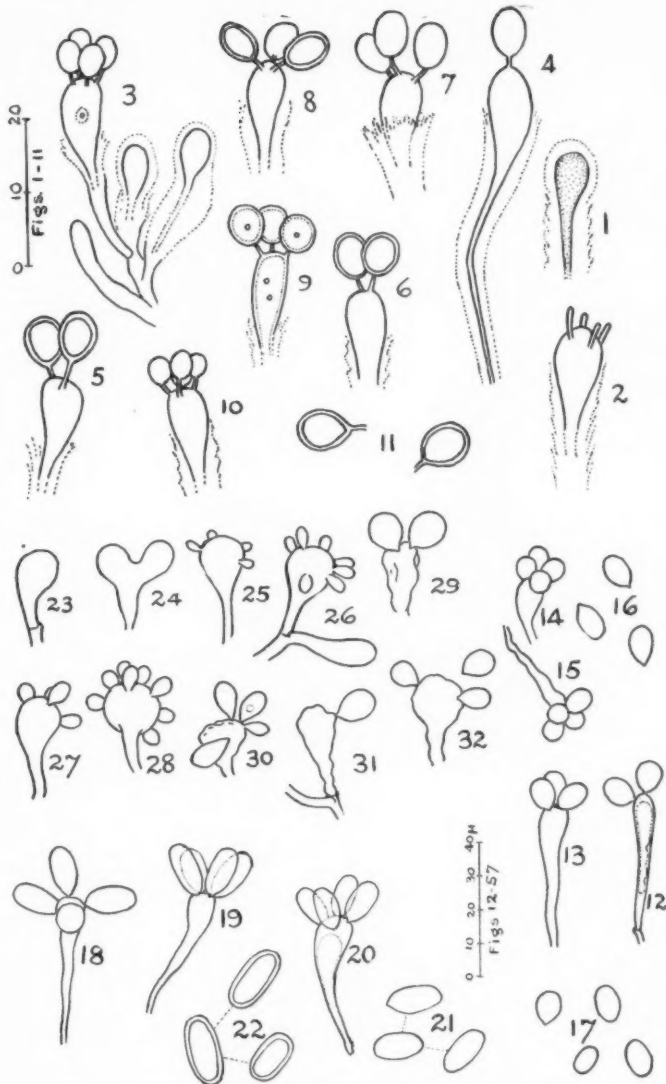
## CYATHUS STERCOREUS

23. Young basidium of usual form; 24. Double basidium; 25-28. Basidia with from four to eight developing spores; 29-31. Basidia collapsing, spores still attached; 32. Collapsing basidium; two spores still attached, one just separated.

## PLATE 23

## CYATHUS STERCOREUS

33-36. Four groups of three spores each from as many mature peridioles, each representing a different collection; 37. Three spores from the outermost peridiole of a newly opened basidiocarp; 38. Three spores from the second peridiole of the same basidiocarp, after ten days in a moist chamber; 39. Nurse hyphae and spore which they had surrounded, separated from them

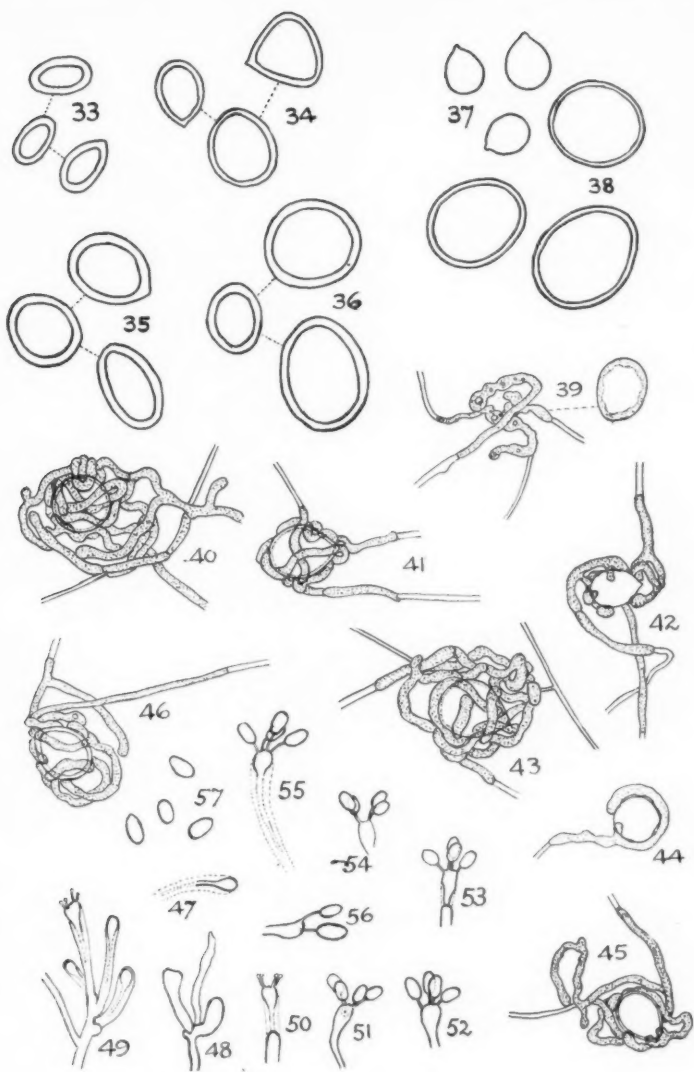


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by pressure on cover slip; 40-46. Spores surrounded by nurse hyphae, the latter somewhat loosened by pressure of cover slip.

CRUCIBULUM VULGARE

47. Young basidium; 48. Group of three young basidia, the middle one of which has become gelatinized and abortive; 49. Cluster of young basidia; 50. Young basidium with spores forming; 51-55. Mature or nearly mature basidia with spores; 56. Abnormal two-spored basidium; one spore unusually large; 57. Mature spores.

# STUDIES OF THE FUNGOUS FLORA OF VIRGIN SOILS<sup>1</sup>

FREDERICK S. PAINE

(WITH PLATES 24-26)

## PART I

### INTRODUCTION

In reviewing the literature on soil fungi and soil bacteria certain tendencies in research in this field are quite noticeable. The greater amount of work has been done with cultivated soils, in the interests of agriculture, rather than with virgin soils; more attention has been given to the physiological reactions of the fungi than to their taxonomy; more definite information seems to have been gained regarding soil bacteria than fungi. Not much has been done in determining the depth at which these fungi occur nor has much been written concerning the relative frequency with which these soil molds occur, and in only a few instances have virgin soils been studied.

Van Iterson (14), in 1904, made a study of what he presumed to be soil fungi, as he obtained them from the air over cultivated fields. In 1907 Hagem began the determination of the Mucors in soil, and in 1910 he (5) reported his findings of various fungi in the soils of Norway. In 1912 Dale (3) made an investigation of the fungi in agricultural soils fertilized with ammonium sulphate with and without lime. In 1914 she (4) made a study of the fungous flora of different kinds of soil including chalky, uncultivated mountain peat, and the "black earth" of reclaimed fenland. Dale's works were largely taxonomic. Jensen (7), in 1912, published his work on the studies of the fungi found in

<sup>1</sup>The author is indebted to the Botany Departments of the State University of Iowa and Iowa State College for the opportunity of doing this research. He wishes to take this opportunity to express his sincere appreciation to Dr. G. W. Martin and Dr. J. C. Gilman, of these Departments, for their very valuable assistance during the progress of this work.



cultivated fields where crop rotation had been practiced; his work also was largely taxonomic. Waksman (15), in 1916, gave a list of 106 species of fungi he had found in garden, orchard, meadow, and forest soils. Abbott (1), in 1926, gave a list of thirty-six species of soil fungi he found in a study of the effects of fertilization on the fungi present in cultivated soils.

The list of references here given is not intended to be complete but is a representative list to show the nature of the work that has been done in this field of study. The work reported in this paper was undertaken in an attempt to obtain some idea of the depth and frequency of the occurrence of these fungi, and also to add to what has already been done in studying the forms inhabiting virgin soils.

#### EXPERIMENTAL WORK

For isolating the fungi two different kinds of media were used, Cook's No. 2 and a beerwort substitute.<sup>2</sup> The beerwort substitute was used in routine work for carrying the stock cultures and growing the fungi for microscopic study.

The soils from which the organisms were isolated were open pasture land that had never been tilled, and timber land, all Clinton silt loam, near Iowa City, Iowa. The isolations were made in late September and early October. Due to a prolonged rainy season at that time the soils were at their saturation points. Samples of soil were taken twice from each kind of land at depths of one inch, three, six, and twelve inches, and once from the surface of the woodland. The samples were collected in small bottles that had been sterilized in the autoclave, and were plated out within eighteen hours after collecting. Dilutions were prepared by weighing out one gram of the wet soil and suspending it in ninety-nine cubic centimeters of sterile tap water; one loopful of this suspension was transferred to five cubic centimeters of sterile tap water, and also to a tube of each kind of medium; the medium was poured into sterile plates, and similarly a loopful from the five-cubic-centimeter

<sup>2</sup> Cook's No. 2 medium was prepared according to the formula given by Brown and Halvorsen (2), and the beerwort substitute was prepared by diluting a bottle of "near beer" (about 350 c.c.) to one liter with tap water, dissolving 15 gms. of agar-agar, filtering through cotton flannel, and sterilizing under pressure.

TABLE I

FREQUENCY AND DEPTH OF SPECIES FOUND IN OPEN PASTURE LAND

Organism	Depth				Organism	Depth			
	1 inch	3 inches	6 inches	Total		1 inch	3 inches	6 inches	Total
<i>Absidia subpoculata</i> .....	1			2	<i>Penicillium chrysogenum</i> .....	1			2
<i>Zygorrhynchus Moelleri</i> ....	1	1		2	<i>Hormodendrum cladosporioides</i> .....	27	3	2	33
<i>Aspergillus fumigatus</i> .....	95	1		96	<i>Hormodendrum olivaceum</i> .....	1			1
<i>Aspergillus versicolor</i> .....	1			1	<i>Acrostalagmus albus</i> .....	1		1	2
<i>Aspergillus terreus</i> .....	1			1	<i>Alternaria</i> No. 1.....	3	1		5
<i>Aspergillus olivaceus</i> .....	1			1	<i>Alternaria</i> No. 2.....	1	1		2
<i>Penicillium decumbens</i> .....			1	1	<i>Alternaria</i> No. 3.....	1	1		2
<i>Penicillium roseum</i> .....	1	1		2	<i>Alternaria</i> No. 5.....	1			1
<i>Penicillium biforme</i> .....	1			1	<i>Fusarium</i> .....	1	1	1	3
<i>Penicillium atramentosum</i> ....		1		1					

TABLE II

FREQUENCY AND DEPTH OF SPECIES FOUND IN WOODLAND

Organism	Depth				Organism	Depth			
	Surface	1 inch	3 inches	Total		Surface	1 inch	3 inches	Total
<i>Mucor hiemalis</i> .....	3		1	4	<i>Spicaria elegans</i> .....		1		1
<i>Mucor Ramannianus</i> .....	1			2	<i>Hormodendrum cladosporioides</i> .....	4	30	2	36
<i>Mucor virus</i> .....		1	1	2	<i>Hormodendrum nigrescens</i> .....	1			1
<i>Mucor echinulatus</i> .....	1			1	<i>Hormodendrum viride</i> .....	2	1		3
<i>Cunninghamella verticillata</i> .....			1	1	<i>Stachybotrys cylindrospora</i> .....			1	1
<i>Chaetomium bostrychodes</i> .....	1	2		3	<i>Trichoderma lignorum</i> .....	2		1	3
<i>Aspergillus nidulans</i> .....	1		1	2	<i>Gliobotrys albo-viridis</i> .....	1			1
<i>Aspergillus niger</i> .....	1			1	<i>Acrostalagmus albus</i> .....	1			1
<i>Aspergillus conicus</i> .....	3			3	<i>Alternaria</i> No. 4.....	1	3		4
<i>Aspergillus Sydowi</i> .....			1	1	<i>Coniothyrium</i> .....			1	1
<i>Penicillium atramentosum</i> .....	2	1		3	<i>Cephalosporium</i> .....	2	1	2	5
<i>Penicillium echinatum</i> .....	2			2					

suspension was inoculated into a tube of each of the different kinds of media and poured into sterile plates. The plates were incubated three days at room temperature. After the three days of incubation the different types of colonies were counted, notes made of the number and depths from which they came, and transfers made to "near beer" agar slants. These findings are recorded in tables I and II.

It would seem, from tables I and II, that *Mucors* are not as abundant in the open pasture land as they are in the forest areas; that *Aspergillus fumigatus* and *Hormodendrum cladosporioides* are very common inhabitants of open pasture soils, particularly near the surface, and also that species of *Alternaria* are frequent inhabitants of such soils. *Hormodendrum cladosporioides* seems also to occur frequently in timber soils. It is interesting to note that the species of *Coniothyrium*, representing a genus commonly parasitic on higher plants, was found six and twelve inches below the surface, and that the *Cunninghamella*, a new species, was also found twelve inches below the surface.

The identification of the species found made up a large part of the problem undertaken. All the supposedly pure cultures were plated out by dilution at least twice, picking each time what seemed to be pure colonies, to minimize the possibility of dealing with mixed cultures. Descriptions of the identified forms follow.

#### DESCRIPTIONS OF SPECIES FOUND

##### PHYCOMYCETES

###### 1. *Absidia subpoculata* n. sp.

Colonies white, floccose, aërial hyphae growing to a height of 1.5 to 2 cm., somewhat spreading. Sporangiophores branched, with shorter sporangiophores borne as lateral branches, one to five, on the main sporangiophore, the branches 100 to 300  $\mu$  long by 4  $\mu$  in diameter with a septum 10 to 12  $\mu$  below the tip. The sporangium has a thick, diffuent wall often leaving a collar attached to the columella; it is globose, 22 to 24  $\mu$  in diameter, quite uniform in size, smooth, columella oval, slightly constricted at the apophysis, 4 to 7  $\mu$  by 8 to 9  $\mu$ . Spores very numerous, oval or spherical to allantoid, 2 to 2.5 by 3 to 4  $\mu$ . Chlamydospores quite numerous, spherical, 4 to 5  $\mu$  in diameter.

Hab. Found in open pasture land 12 in. below the surface.

## 2. *MUCOR HIEMALIS* Wehmer.

Colonies white, floccose, 0.5 to 2 cm. high. Sporangiophores hyaline, unbranched, 1.5 or more cm. long by 5 to 8  $\mu$  thick, arising singly from the substratum. Sporangia dark, spherical to subglobose, smooth, deliquescent when old, 65 to 75  $\mu$  in diameter. Columella 35 to 40  $\mu$  in diameter, oval, somewhat flattened at the base. Spores round, hyaline (smoky in mass), 8 to 11  $\mu$  in diameter. Chlamydospores not conspicuous, somewhat irregular in form and size.

Hab. Found in timber land from the surface to 6 in. below the surface.

## 3. *MUCOR RAMANNIANUS* Moeller.

Colonies somewhat spreading, zonate, often papillate near the center, velvety, rusty red in color. Sporangiophores hyaline, unbranched, 50 to 100  $\mu$  long by 4  $\mu$  thick, arising in large numbers from the substratum. The sporangia spherical, quite uniform in size, 13 to 14  $\mu$  in diameter, deliquescent. Columella spherical, 5 to 6  $\mu$  in diameter. Chlamydospores appear as irregular oval or spherical swellings in the vegetative hyphae of the substratum.

Hab. Found in timber soil on the surface and 12 in. below the surface.

## 4. *Mucor mirus* n. sp.

Colonies flat, except that they are usually papillate in the center, zonate, quite spreading, velvety, ashy gray, scarcely any elevation. Sporangiophores unbranched, arising in the margin of the colony in large numbers but not numerous inside the margin, rather delicate and slender, 125 to 300  $\mu$  long and 2 to 2.5  $\mu$  thick. Sporangia smoky, spherical, 10 to 30  $\mu$  in diameter, appearing slightly rough; the sporangial wall is deliquescent, leaving no collar. Columella spherical, 5 to 12  $\mu$  in diameter. Spores small, 2.5 to 3.5  $\mu$  in diameter, often oval and slightly pointed at the ends. Chlamydospores are exceedingly numerous, and appear as large globular swellings in the subterranean hyphae, 20 to 35  $\mu$  in diameter, and appear to contain from 2 to 8 angular spore-like bodies varying from 4 to 12  $\mu$  across, rather angular in shape.

Hab. Found in timber soil 1 to 6 in. from the surface.

This form, except for its color and habit of spreading widely, closely resembles *M. Ramannianus* in its macroscopic appearance, but differs microscopically from the latter in its long slim sporangiophore, bearing the sporangiophores mostly near the

margin of the colony, very small oval spores, and the abundance and appearance of the chlamydospores.

5. *Mucor echinulatus* n. sp.

Colonies somewhat spreading, soon developing a grayish white powdery appearance; elevation of the aerial hyphae never exceeding 2 mm. in height; turf prostrate, fragile. Sporangio-phores monopodially branched, 150 to 400  $\mu$  in length by 5.5 to 8  $\mu$  thick, much vacuolated. Sporangia spherical, 40 to 50  $\mu$  in diameter, minutely echinulate, the spines being about 2  $\mu$  long; the sporangial wall deliquescent, leaving no collar. Columella spherical, quite variable in size, 15 to 25  $\mu$  in diameter. Spores ellipsoidal, 3 to 3.5  $\mu$  by 4.5 to 5.5  $\mu$ , hyaline. Chlamydospores quite numerous in old cultures, cylindrical in form, quite variable in length.

Hab. Found in the surface soil of timber land.

This species somewhat resembles *M. dispersus*, but differs from that species in scanty growth, its globose columella, and its much smaller spores.

6. *ZYGORRHYNCHUS MOELLERI* Vuillemin.

Colonies spreading rapidly, white, becoming ashy with age; aerial hyphae arising 1.5 to 2 cm. Zygosporangia numerous, rough, warty, very dark when mature, 30 to 45  $\mu$  in diameter; the larger suspensor 20 to 30  $\mu$  thick at its thickest point. Sporangio-phores slender, 1 or more mm. in length and 6 to 8  $\mu$  thick. Sporangia dark, rough, nearly spherical, 40 to 50  $\mu$  in diameter, usually rupturing at the apex. Columella somewhat flattened, oval, 20 to 27  $\mu$  by 25 to 35  $\mu$ . Spores ovoid to ellipsoidal, 2.5 to 3.5  $\mu$  by 4 to 7  $\mu$ .

Hab. Found in open pasture land 1 to 6 in. below the surface.

7. *Cunninghamella verticillata* n. sp.

Colonies spreading; aerial hyphae loose, elevated, 2 to 4 cm. in height, somewhat silvery, much vacuolated. Conidiophores very long, 2 cm. or more, and 12 to 14  $\mu$  in thickness. Numerous lateral branches are borne at various places along the conidiophore just below the terminal vesicle, forming a number of whorls of two to six lateral branches, each terminating in a vesicle, the conidiophore being more or less swollen at each point of attachment of the lateral branches; lateral branches not exceeding 30  $\mu$  in length, their vesicles pyriform or oval in shape, not over 16  $\mu$  in diameter. The terminal vesicle globose to oval, about

50  $\mu$  in diameter. Spores borne on the terminal vesicle ellipsoidal, pointed at the attached end, 10  $\mu$  by 13 to 15  $\mu$ ; spores borne on the lateral vesicles oval, bluntly pointed at the attached end, 8 to 12  $\mu$  by 12 to 15  $\mu$  in diameter. All spores are adorned with fine echinulations, 1.5 to 3  $\mu$  in length.

Hab. Found 12 in. below the surface in timber land.

This species differs from *C. echinulata* in size of spores, in leaving scarcely any points on the vesicles where the spores were attached, and in the appearance of the turf.

#### ASCOMYCETES

##### 8. CHAETOMIUM BOSTRYCHODES Zopf.

Colony formed of sterile creeping hyphae with scarcely any elevation, at first white, later becoming brown. Perithecia quite evenly scattered over the surface of the substratum, dark olive green, 150 to 250  $\mu$  in diameter, adorned with many unbranched appendages 300 to 500  $\mu$  long tapering to a point; those from the upper part of the perithecium having from 4 to 6 spiral coils, each coil 20 to 40  $\mu$  long by 24 to 28  $\mu$  wide, the lower appendages being nearly straight. The interior of the perithecium is filled with many asci; asci clavate and borne on a rather long stipe; asci 12 to 15  $\mu$  thick by 20 to 30  $\mu$  long, the stipe 15 or more  $\mu$  long, total length 45 to 60  $\mu$ . Asci usually contain 8 spores in 2 ranks. Ascospores subspherical, 7 to 8  $\mu$  by 9 to 10  $\mu$ , slightly pointed at each end.

Hab. Isolated from open pasture land 1 to 3 in. below the surface.

#### FUNGI IMPERFECTI

##### 9. ASPERGILLUS FUMIGATUS Fresenius.

Colonies grayish blue, moderate elevation. Mycelium hyaline, branched. Conidiophores 150 to 300  $\mu$  long by 4 to 6  $\mu$  thick; vesicle globose to flask-shaped, 15 to 20  $\mu$  in diameter; conidia green, borne in large columnar masses, smooth, round, 2.5 to 3.5  $\mu$  in diameter, attached by 1-seriate sterigmata; sterigmata 5 to 6  $\mu$  long and somewhat pointed at the tip.

Hab. Found in pasture land soil in large numbers 1 in. from the surface and rarely 6 in. below the surface.

##### 10. ASPERGILLUS VERSICOLOR Vuillemin.

Colony velvety, greenish blue; reverse tinged with olive green. Hyphae slightly smoky. Conidiophores unbranched, hyaline, 50

to  $250\ \mu$  long by  $3.5$  to  $4\ \mu$  in diameter; vesicle pyriform,  $10$  to  $12\ \mu$  in diameter; sterigmata 2-seriate,  $10$  to  $12\ \mu$  long, originating mostly at the apex of the vesicle, giving somewhat the appearance of a tuft of bristles; conidia round, greenish blue, smooth,  $3$  to  $4\ \mu$  in diameter.

Hab. Found in pasture land soil  $3$  in. below the surface; not numerous.

#### 11. *ASPERGILLUS TERREUS* Thom.

Colonies at first white, later becoming ochraceous; compact or velvety; conidiophores short,  $80$  to  $140\ \mu$  long by  $3.5$  to  $5\ \mu$  thick; vesicle globose to flask-shaped,  $12.5$  to  $15\ \mu$  in diameter; sterigmata 2-seriate, the primary  $6$  to  $7\ \mu$  long, the secondary  $5$  to  $6\ \mu$  long; conidia globose, smooth, hyaline or nearly so,  $2.5$  to  $3\ \mu$  in diameter.

Hab. Found in open pasture land  $1$  in. below the surface; not numerous.

#### 12. *ASPERGILLUS NIDULANS* Eidam.

Colonies small and velvety, blue-green. Conidiophores short,  $100$  to  $150\ \mu$  by  $4\ \mu$ ; vesicle ovate,  $10$  to  $12\ \mu$  in diameter; sterigmata 2-seriate, short and thick, total length  $7$  to  $9\ \mu$ . Conidia borne in moderately long chains, green, smooth, spherical to subspherical,  $3$  to  $3.5\ \mu$  in diameter; the conidial mass forming a column.

Hab. Found in woodland soil  $1$  and  $12$  in. from the surface; not numerous.

#### 13. *ASPERGILLUS NIGER* Van Tieghem.

Young colonies straw colored, finally becoming black as if dusted with a black powder; somewhat spreading, elevation  $1$  to  $2$  mm. Conidiophores long and straight; vesicle globose,  $34$  to  $38\ \mu$  in diameter, bearing a mass of conidia that is nearly spherical in outline. Sterigmata 2-seriate, the primary  $6$  to  $12\ \mu$  long, the secondary  $5$  to  $9.5\ \mu$  long. Conidia borne in moderately long chains, dark, rough, spherical or nearly so,  $3.5$  to  $4.5\ \mu$  in diameter.

Hab. Isolated from the surface soil of timber land.

#### 14. *ASPERGILLUS CONICUS* Blochwitz.

Colonies small, wart-like, grayish blue; reverse cream colored to purplish. Conidiophores  $200$  to  $260\ \mu$  in length by  $6\ \mu$  in thickness; vesicle  $10\ \mu$ , or less, in diameter, ovoid. Sterigmata

1-seriate, borne on the upper half of the vesicle only, and few to moderately abundant, sometimes only a single sterigmatum with a single chain of conidia. Conidia minutely spiny to verrucose, globose to subglobose,  $4\ \mu$  in diameter.

Hab. Isolated from 6 in. below the surface of forest soil.

15. *ASPERGILLUS SYDOWI* Bainier & Sartory.

Colonies small, colorless when young, becoming blue-green; surface velvety, rarely if ever becoming floccose; margin very narrow and almost colorless; reverse creamy white. Conidiophores 150 to  $250\ \mu$  long by 4 to  $5\ \mu$  thick, often branched, bearing conidial heads with subspherical vesicles, 10 or more  $\mu$  in diameter, while others bear few to a single sterigma with a single chain of conidia. Sterigmata 2-seriate except where borne singly as lateral branches, often several secondary sterigmata borne on one primary. Primary sterigmata  $6\ \mu$  long; secondary sterigmata  $4.5$  to  $5\ \mu$  long. Conidia green, smooth, globose,  $4\ \mu$  in diameter.

Hab. Found in the surface soil of forest land.

16. *ASPERGILLUS* (*Citromyces*) *OLIVACEUS* Delacroix.

Colonies green, slightly conical in the fruiting area, with a tendency to become slightly floccose with age, bordered by a hyaline margin 2 to 4 mm. wide; reverse ashy pale green to pale yellow surrounded by a hyaline margin of subterranean hyphae. Conidiophores branched, with numerous lateral branches, 50 to  $100\ \mu$ , or more, in length,  $3.5$  to  $4\ \mu$  in diameter, more or less bent, terminating in a tuft of 1-seriate sterigmata; sterigmata 3 to 8 in number, flask-shaped, 11 to  $15\ \mu$  long by 4 to  $4.5\ \mu$  thick, with a constricted tip. Conidia globose, 4 to  $4.5\ \mu$  in diameter, smooth.

This form differs from *A. Amstelodami* (Mangin) in having smooth instead of echinulate spores, and no perithecia were demonstrated.

Hab. Found 1 in. below the surface of open pasture land.

17. *PENICILLIUM DECUMBENS* Thom.

Colonies greenish blue, only slightly elevated, margin narrow, zonate; reverse greenish yellow. Conidiophores 200 or more  $\mu$  in length by  $3.5\ \mu$  in diameter. Conidia borne in long chains which are twisted together like the strands of a rope, ellipsoidal to oval,  $2.5$  to  $3\ \mu$  by  $5$  to  $6\ \mu$ , greenish blue, fairly uniform in



size and shape. Odor, none. Gelatin liquefied in 12 days, rather viscous, and colored.

Hab. Isolated from 12 in. below the surface of open pasture land.

18. *PENICILLIUM ROSEUM* Link (?).

Colonies at first white, slightly floccose, becoming pink with age; spreading, zonate; reverse white. Conidiophores straight, coarse or fine, quite variable in length and diameter, may be a very short branch of a sterile hypha or a very long coarse stalk, 50 to 300  $\mu$ , or more, in length, by 2.5 to 4.5  $\mu$  in diameter. Conidia quite variable in size, more often fusiform, 2 to 3  $\mu$  by 2.5 to 3.5  $\mu$ , hyaline. Odor, slightly moldy. Gelatin liquefied in 6 days and colored yellow.

Hab. Found 1 in. below the surface of open pasture land.

19. *PENICILLIUM BIFORME* Thom.

Young growth white, becoming grayish with age; surface closely strict, occasionally with sterile hyaline mycelium overgrowing the conidiophores; margin narrow, usually light in color, zonate; reverse creamy white. Conidiophores branched, 75 to 150  $\mu$  long by 3.5 to 4  $\mu$  in thickness. Conidia blue, ovate to spherical, 3 to 4.5  $\mu$  in diameter, borne on flask-shaped sterigmata, with 2 to 8 in a chain. Odor, strong musty. Gelatin liquefied in 12 days without coloring the medium.

Hab. Isolated from 1 in. below the surface of open pasture land.

20. *PENICILLIUM ATRAMENTOSUM* Thom.

Colonies light green with a tendency to become whitish with age, compact with central elevation and tendency to zonation; margin hyaline and narrow, 0.5 to 1 mm. wide; reverse greenish yellow to orange. Conidiophores 250 to 400  $\mu$  long by 4  $\mu$  in thickness, unbranched except at the apex. Conidia somewhat angular, 2.5 to 3  $\mu$  in diameter, subspherical, green. Odor, none. Gelatin liquefied in 6 days without coloring the medium.

Hab. Found 6 in. below the surface of open pasture land.

21. *PENICILLIUM CHRYSOGENUM* Thom.

Colonies grayish blue, flat, with granular surface, spreading; reverse greenish yellow. Conidiophores septate, unbranched except at the apex, 100 to 400  $\mu$  in length by 4 to 4.5  $\mu$  in thickness. Conidia round, 3.5 to 4  $\mu$  in diameter, smooth. Odor, none. Gelatin rapidly liquefied and becoming yellowish.

Hab. Isolated from 3 in. below the surface of open pasture land.

22. *PENICILLIUM ECHINATUM* Dale.<sup>3</sup>

Colonies small to medium sized, blue gray, velvety surface, only slightly elevated, margin narrow; reverse soon becoming a deep purple or wine colored. Conidiophores branched, septate, pigmented, varying in length from short lateral branches  $20\ \mu$  long to a main conidiophore 300 or more  $\mu$  in length, 3.5 to  $5\ \mu$  thick. Conidia verrucose, angular, orange colored, 3 to  $3.5\ \mu$  in diameter, borne in chains of 20 or more. Odor, none. Gelatin rapidly liquefied without coloring the medium.

Hab. Isolated from 1 in. below the surface of timber land.

23. *SPICARIA ELEGANS* Corda.

Colonies white, floccose above and having a powdery appearance beneath. Fertile hyphae borne irregularly on the vegetative hyphae, distinctly septate, branched; the branches numerous and borne in whorls. Conidiophores short, erect, and adorned with short stout bristles. Conidia borne on flask-shaped sterigmata, in chains of 20 or more, fusiform, hyaline,  $2.5$  to  $4\ \mu$  by  $5$  to  $6\ \mu$ .

Hab. Isolated from timber land soil 1 in. from the surface.

24. *HORMODENDRUM CLADOSPORIOIDES* Fresenius.

Colonies flat, dark olive green, spreading. Conidiophores short, septate, smoky, arising from the sterile creeping hyphae, branched,  $150$  to  $200\ \mu$  long by  $4\ \mu$  thick. Conidia dark green, 1-celled, the chains often branching, quite variable in size, oval,  $2$  to  $3\ \mu$  by  $3$  to  $5\ \mu$  for those borne at the tips.

Hab. Found many times from 1 to 3 in. below the surface of both open pasture and timber land, and occasionally 6 in. below the surface of woodland soils.

25. *HORMODENDRUM OLIVACEUM* Corda.

Colonies olive green, slightly elevated, not spreading much; margin narrow, 1 to  $1.5\ \text{mm.}$ , hyaline; reverse dark green. Sterile hyphae soon growing beyond the conidiophores as the colony matures. Conidiophores short,  $75$  to  $200\ \mu$  long, borne as short lateral branches of the sterile hyphae, unbranched except at the apex. Conidia ellipsoidal to short cylindric,  $4$  to

<sup>3</sup> *P. echinatum* Dale was identified by Miss Margaret Church, U. S. Department of Agriculture, Bureau of Animal Industry, Washington, D. C.

5  $\mu$  by 8 to 12  $\mu$ , with intermediate cells of the chain swollen, and terminal cells often much smaller, and globose.

Hab. Isolated from pasture land 1 in. below the surface.

26. *Hormodendrum nigrescens* n. sp.

Colonies somewhat elevated, at first hyaline, becoming olive green, and finally black beneath a white surface; hyaline mycelium appearing slightly floccose; margin 2 or more mm. wide, hyaline. Sterile hyphae arising at the apex of the colony as fine bristle-like tufts above the conidiophores. Conidiophores originating in the substratum, smoky, 300 to 400  $\mu$  long by 4.5 to 5  $\mu$  in thickness, dendroidally branched, erect. Conidia green, subspherical to ellipsoidal or fusiform, though seldom pointed at the ends, 4 to 10  $\mu$  by 2.5 to 4  $\mu$ .

Hab. Found in timber land soil, on the surface and 6 in. below the surface.

This form differs from *H. olivaceum*, which it resembles, in shape and size of conidia; in the origin and length of the conidiophores; and in the coarseness and abundance of the sterile hyphae.

27. *HORMODENDRUM VIRIDE* Fresenius.

Colonies somewhat floccose above an olive green, somewhat restricted growth of creeping hyphae. Conidiophores short, dendroidally branched. Conidia yellowish green, very irregular in shape and size, oval to cylindrical, sometimes indistinctly guttulated, smooth, 4 to 12  $\mu$  by 2 to 4  $\mu$ .

Hab. Found in the surface soil of timber land.

28. *STACHYBOTRYS CYLINDROSPORA* Jensen.

Colonies spreading, somewhat zonate, becoming sooty with age. Sterile hyphae hyaline, prostrate, creeping, branched. Conidiophores unbranched, hyaline at the base, becoming fuliginous near the apex, and tapering, 65 to 85  $\mu$  in length by 3 to 3.5  $\mu$  in thickness; sterigmata 4 to 9 in number, borne at the apex of the conidiophore, 11  $\mu$  by 4 to 5  $\mu$ . Conidia black, easily detached from the sterigmata, ellipsoidal, 1-celled, 2.5 to 4  $\mu$  by 5 to 10  $\mu$ .

Hab. Isolated from soil 6 in. below the surface of timber land.

29. *TRICHODERMA LIGNORUM* Tode.

Colonies of a scanty spreading growth of hyaline mycelium, forming in 3 to 5 days green cushion-like structures of fruiting

masses. Conidiophores verticillately branched. Conidia green, spherical or nearly so, 1-celled,  $2.5$  to  $3\ \mu$  in diameter.

Hab. Found in woodland soil from the surface to 12 in. below the surface.

30. *GLIOBOTRYS ALBO-VIRIDIS* von Höhnelt.

Colonies flat and spreading; the sterile hyphae very crooked, septate,  $3$  to  $7.5\ \mu$  in thickness, profusely branched, greenish colored; aerial hyphae bearing lateral branches of fertile hyphae. The fertile hyphae branched, each branch terminating in a spherical envelope filled with globular spores. When young and immature a slimy capsule appears to surround the adjacent young envelopes of the secondary branches. Conidiophores smoky,  $20$  to  $30\ \mu$  long by  $3\ \mu$  thick, verticillately branched at the tips, usually 3 branches, the terminal branches  $9$  to  $14\ \mu$  long by  $3\ \mu$  in thickness. Conidia borne in slime, globose to ovoid,  $3.5$  to  $4\ \mu$  in diameter, smooth, green. Many chlamydospores are borne in the substratum.

Hab. Isolated from the surface soil of timber land.

31. *ACROSTALAGMUS ALBUS* Preuss.

Colonies white, very floccose, spreading, elevated; mycelium septate. Conidiophores more or less septate near the base, rather crooked,  $50$  to  $150\ \mu$  long by  $4\ \mu$  thick, often profusely branched near the tip; branches in whorls of 2 to 4, usually 3. Sterigmata somewhat club-shaped, tapering toward the tip,  $10$  to  $13\ \mu$  long, in whorls of 2 to 4 usually. Conidia ellipsoidal to crescentiform or allantoid, hyaline, smooth,  $4$  to  $9$  (rarely  $9$ )  $\mu$  by  $2.5$  to  $4\ \mu$ .

Hab. Found in open pasture land 1 to 12 in. below the surface, and on the surface of timber land.

Among the several species of fungi found there were five distinct species of *Alternaria*, the identification of which was a greater problem than could be included in this work. They were all frequently found and have likely been seen by other investigators. They are reported here by number and briefly described.

32. *ALTERNARIA* No. 1.

Colonies somewhat spreading, with a margin 2 to 3 mm. wide, dark olive green; aerial hyphae extending up to 10 to 15 mm., floccose, smoky to nearly hyaline, dense. Subterranean

hyphae dark olive green, straight. Conidiophores numerous, short, septate, straight, often more or less constricted, dark, 4 to 5  $\mu$  in diameter. Conidia smooth, usually 3- to 8-celled, 12 to 36  $\mu$  long by 6 to 12  $\mu$  broad, moderately muriform, brown, borne in chains of 2 to 10 usually.

Hab. Isolated from open pasture land 1 to 6 in. below the surface.

### 33. *ALTERNARIA* No. 2.

Colonies a sooty green, becoming almost black; growth 3 to 5 mm. high; no margin; distinctly zonate, with a dark central area surrounded by a light zone surrounded again by a dark zone which in turn is bordered by a second light zone; aerial hyphae short, somewhat scanty, wooly and smoky; submerged mycelium blackish green, straight; conidiophores abundant, short, branched, dark, often constricted, 3.5 to 4.5  $\mu$  in diameter. Conidia very dark, borne in long chains, often as many as 30 in a chain, strongly muriform, smooth, 2 to 12 cells, 12 to 30  $\mu$  long by 6 to 16  $\mu$  thick.

Hab. Found in open pasture land from 1 to 6 in. below the surface.

### 34. *ALTERNARIA* No. 3.

Colonies spreading, zonate, with a smoky central area, surrounded by a wide light zone bordered by a very narrow dark band and this in turn surrounded by a wider greenish gray zone which blends off into a very narrow hyaline margin. Growth 5 to 8 mm. high; aerial mycelium cottony, pale greenish gray; reverse, concentric rings of light and dark olive green; submerged mycelium olive green, nearly straight. Conidiophores numerous, dark, very short, much constricted, septate. Conidia numerous, borne in long chains, olive brown, smooth, 2- to 14-celled, scantily muriform, 6 to 12  $\mu$  by 10 to 20  $\mu$ .

Hab. Found in open pasture land 1 to 6 in. below the surface.

### 35. *ALTERNARIA* No. 4.

Colonies spreading, zonate with a dark central area surrounded by a lighter area, then a grayish wooly zone blending into a hyaline margin 1 to 2 mm. wide. Aerial hyphae not very abundant, 4 to 6 mm. high, ashy gray; submerged mycelium nearly straight, dark olive green, moderately branched; conidiophores very numerous, normal or only slightly constricted, dark green, 40 to 50  $\mu$  long by 6 to 7  $\mu$  thick. Conidia very numerous,

borne in long chains, often containing many cells, rough, muriform, dark olive brown, 18 to 32  $\mu$  long by 7 to 14  $\mu$  in thickness.

Hab. Isolated from timber land soil 1 to 6 in. below the surface.

#### 36. ALTERNARIA No. 5.

Colonies without zonation, no pronounced spreading, no margin; aerial hyphae abundant, grayish green, wooly; submerged mycelium dark olive green, straight. Conidiophores scanty, short, many septa, constricted, 5 or more  $\mu$  in thickness. Conidia not numerous, borne in short chains, rarely more than 5 or 6 in a chain, short, thick, profusely muriform, olive green.

Hab. Isolated from open pasture land 1 in. from the surface.

#### 37. CONIOTHYRIUM (species not identified).

Pycnidia borne more or less scattered on the surface of the medium as dark brown specks; measurements from a fixed section, 100 to 160  $\mu$ . The surface of the medium covered with a thin mat of sterile creeping mycelium. Spores borne in the pycnidium in large numbers, purple, ovate, 2.5 to 3  $\mu$  by 4 to 5  $\mu$ .

Hab. Isolated from the soil of timber land 6 and 12 in. below the surface.

## PART II

### CELLULOSE FERMENTATION

#### INTRODUCTION

Of the various physiological reactions of soil fungi their action on cellulose has been studied as one of the most important. As early as 1902 Omelianski (11) demonstrated the ability of certain soil bacteria to ferment cellulose. Others soon took up the study of cellulose fermentation. In 1904 Van Iterson (14), during his work of isolating air fungi, showed that many of them could cause cellulose decay. In 1912 Kellerman and McBeth (9) published a formula for preparing cellulose agar medium on which to test cellulose digestion. Since then this medium, or a modification of it, has been generally used in cellulose fermentation studies. The medium<sup>4</sup> used in this work differs from that

<sup>4</sup> Note: The medium used in this work was made up as follows: one liter of distilled water, four grams of ammonium phosphate, two grams of dipotassium phosphate, and two tenths of one gram of magnesium sulphate.

reported by previous workers in that all sources of carbon are eliminated except that in the cellulose material and that in the traces of carbon dioxide in the air. With cellulose agar Kellerman (8), in 1913, showed the excretion of cytase by *Penicillium pinophilum*. In 1915 Scales (12) gave a list of thirty-one species of cellulose fermenting fungi, but these were not nearly all soil forms. Murray (10), in 1921, reported his findings on the effects of different amounts of straw, used as fertilizer, on the total and available nitrogen in such fertilized soils. In 1924 Starkey (13) gave the results of his study on the rate of decomposition of different kinds of organic material (dried blood, mold mycelium, alfalfa meal, rye straw, and dextrose) and their effects on the amount of available nitrogen. Heukelekian and Waksman (6), in 1925, used *Trichoderma Koningi* and a *Penicillium* in a study of the cycle of carbon from decaying cellulose.

#### EXPERIMENTAL WORK

The fungi isolated in this research were inoculated into the different cellulose media and incubated at room temperature until good growth appeared, or until the thirty-fifth day when there was no growth before that time. Growth was determined by the discoloration and disintegration of the cellulose material. Organisms that failed to grow on the cotton were not cultured on filter paper. The results are shown in table III. Positive results are shown by a (+) and negative results are shown by a (-); where no test was made the place is left blank.

From table III it would appear that the *Phycomycetes* are not active fermenters of cellulose or the medium used was not suitable for their growth. Of the *Aspergilli* and the *Penicillii* it appears that many of them are able to attack cellulose though a goodly number of them cannot. Among the active cellulose destroyers of the soil may be named species of *Hormodendrum*, *Stachybotrys*, *Gliobotrys*, *Acrostalagmus*, *Alternaria*, *Fusarium*, and *Cephalosporium*.

A small amount of the cellulose material was put in a culture tube and enough of the solution was added to cover about two thirds of the cellulose material. The tubes were then sterilized under fifteen pounds of steam pressure. The cellulose materials used were cotton batting, absorbent cotton, and ashless filter paper.

TABLE III  
CELLULOSE FERMENTATION

Organism	Source	Cotton batting	Absorbent cotton	Filter paper	Organism	Source	Cotton batting	Absorbent cotton	Filter paper
<i>Absidia subpoculata</i> ..	Pasture	-	-	-	<i>Penicillium chrysogenum</i> ....	Pasture	+	+	-
<i>Mucor hiemalis</i> .....	Woods	-	-	-	<i>Penicillium echinatum</i> .....	Woods	+	+	-
<i>Mucor Ramanianus</i> .....	"	-	-	-	<i>Fusarium</i> .....	Pasture	+	+	+
<i>Mucor mirus</i> .....	"	-	-	-	<i>Spicaria elegans</i> ....	"	-	-	-
<i>Mucor echinulatus</i> ...	"	-	-	-	<i>Hormodendrum cladosporioides</i> ..	"	+	+	+
<i>Zygorrhynchus Moelleri</i> .....	Pasture	-	-	-	<i>Hormodendrum olivaceum</i> .....	"	+	+	+
<i>Cunninghamella verticillata</i> .....	Woods	-	-	-	<i>Hormodendrum nigrescens</i> .....	Woods	-	+	+
<i>Chaetomium bostrychodes</i> .....	"	-	+	+	<i>Hormodendrum viride</i> .....	"	-	-	-
<i>Aspergillus fumigatus</i> .....	Pasture	+	+	+	<i>Stachybotrys cylindrospora</i> ....	"	+	+	+
<i>Aspergillus Tiraboschii</i> .....	"	+	+	+	<i>Trichoderma lignorum</i> .....	"	+	+	-
<i>Aspergillus terreus</i> ...	"	+	+	+	<i>Gliobotrys albobiridis</i> .....	"	+	+	+
<i>Aspergillus nidulans</i> ...	Woods	-	-	-	<i>Acrostalagmus albus</i> .....	Pasture	+	+	+
<i>Aspergillus niger</i> ....	"	-	-	-	<i>Alternaria</i> No. 1....	"	+	+	+
<i>Aspergillus conicus</i> ..	"	-	-	-	<i>Alternaria</i> No. 2....	"	+	+	+
<i>Aspergillus Sydowi</i> ..	"	+	+	+	<i>Alternaria</i> No. 3....	"	+	+	+
<i>Aspergillus olivaceus</i> ..	Pasture	-	-	-	<i>Alternaria</i> No. 4....	"	+	+	+
<i>Penicillium decumbens</i> .....	"	+	+	+	<i>Alternaria</i> No. 5....	Woods	+	+	+
<i>Penicillium roseum</i> ...	"	-	-	-	<i>Coniothyrium</i> .....	"	-	-	-
<i>Penicillium atramentosum</i> .....	Woods and Pasture	+	+	-	<i>Cephalosporium</i> ....	"	+	+	+
<i>Penicillium biforme</i> ..	Pasture	-	-	-					

## SUMMARY

In this survey of virgin soils thirty species of soil fungi were found and identified, five of which are described as new species, viz., *Absidia subpoculata*, *Mucor mirus*, *Mucor echinulatus*, *Cunninghamella verticillata*, and *Hormodendrum nigrescens*. Of five species of *Mucor* found two were new species; there were five



species of *Hormodendrum*, one of which was new; there were eight species of *Aspergillus* found, one of which, *A. fumigatus*, occurred in large numbers; there were found seven species of *Penicillium*; one species of *Coniothyrium*, unidentified, was found, once six and once twelve inches below the surface. Of the thirty identified species only three, *Penicillium atramentosum*, *Hormodendrum cladosporioides* and *Acrostalagmus albus*, were found common to both woodland and open pasture land.

Many of the species were found capable of growing on a synthetic medium with cellulose as the only source of carbon. Half the species of *Aspergillus* could ferment cellulose, while five of the species of *Penicillium*, of the seven found, showed cellulose-fermenting power to a considerable degree. The species of *Hormodendrum* and *Alternaria* seem to be almost constant in their ability to ferment cellulose.

There seems to be a marked diminution in the frequency of occurrence of fungi with increase in depth beyond the first three inches, in virgin soils, but the lower depths yield their proportion of the new species; four of the five new forms found were isolated from six to twelve inches below the surface.

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## PLATE 24

1. *Absidia subpoculata*: a, sporangium,  $\times 700$ ; b, columella,  $\times 700$ ; c, branching of sporangium, and septum near the tip and bearing a columella with a collar remaining,  $\times 700$ ; d, spores,  $\times 1400$ ; e, young sporangiophore and sporangium developing,  $\times 700$ .

2. *Mucor mirus*: a, sporangiophore and sporangium,  $\times 1400$ ; b, columella,  $\times 1400$ ; c, spore,  $\times 1400$ ; d, chlamydospores,  $\times 700$ .

3. *Mucor echinulata*: a, sporangiophore showing monopodial branching,  $\times 450$ ; b, sporangium,  $\times 450$ ; c, columella,  $\times 700$ ; d, chlamydospore,  $\times 700$ .

4. *Cunninghamella verticillata*: a, upper end of the conidiophore showing the terminal vesicle,  $\times 350$ ; b, portion of the conidiophore showing the entire system of lateral branches,  $\times 350$ ; c, lateral branch with spores attached,  $\times 450$ ; d, spore from the terminal vesicle,  $\times 900$ ; e, spore from a lateral vesicle,  $\times 900$ .

5. *Hormodendrum nigrescens*: conidial head and spores,  $\times 700$ .

6. *Mucor hiemalis*: a, sporangium,  $\times 700$ ; b, columella showing some of the basal collar,  $\times 450$ .

7. *Mucor Ramannianus*: a, sporangium and sporangiophore, showing the thick stout sporangiophore,  $\times 1400$ ; b, columella with some of the basal collar,  $\times 1400$ ; c, spores showing the angular appearance,  $\times 1400$ ; d, chlamydospores,  $\times 700$ .

8. *Zygorrhynchus Moelleri*: a, sporangiophore,  $\times 700$ ; b, columella,  $\times 700$ ; c, mature zygosporangium,  $\times 450$ ; d, developing zygosporangium,  $\times 450$ .

9. *Chaetomium bostrychodes*: a, diagrammatic sketch of the perithecium, showing the asci; b, asci,  $\times 450$ ; c, spores,  $\times 700$ .

10. *Aspergillus versicolor*: vesicle, sterigmata, and spores detached,  $\times 700$ .

## PLATE 25

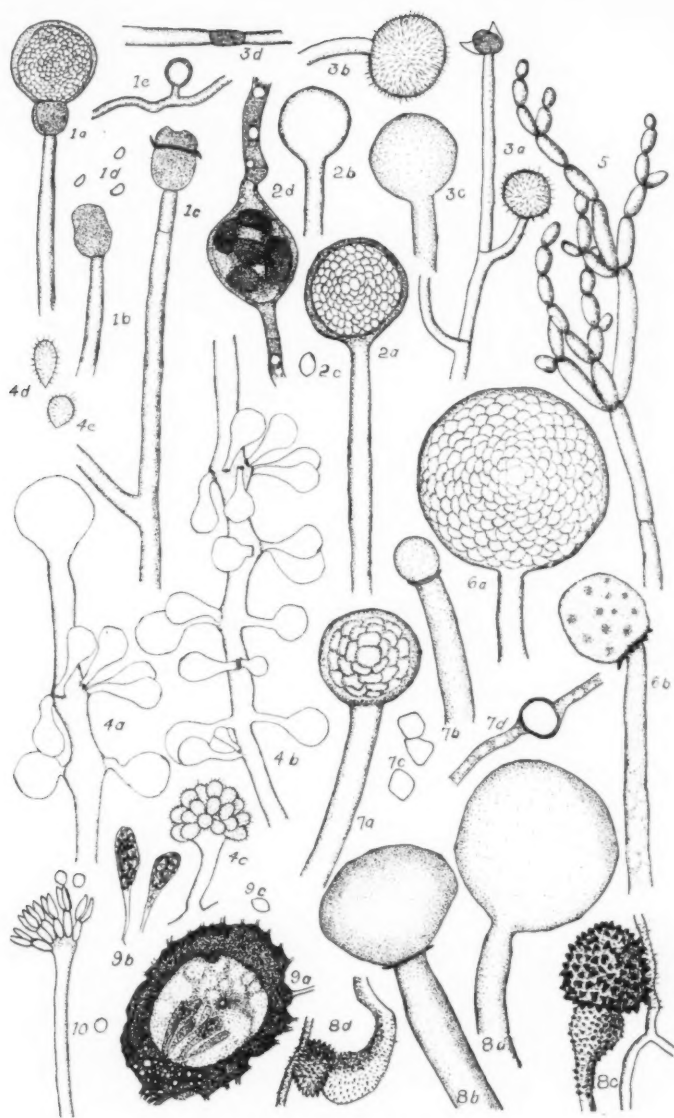
11. *Aspergillus fumigatus*: a, upper part of conidiophore, vesicle, and sterigmata,  $\times 700$ ; b, diagrammatic sketch of the conidial head showing columnar appearance,  $\times 450$ .

12. *Aspergillus terreus*: a, lateral branching of hyphae,  $\times 450$ ; b, diagrammatic sketch of vesicle and sterigmata with chains of conidia,  $\times 700$ .

13. *Aspergillus nidulans*: vesicle and sterigmata,  $\times 700$ .

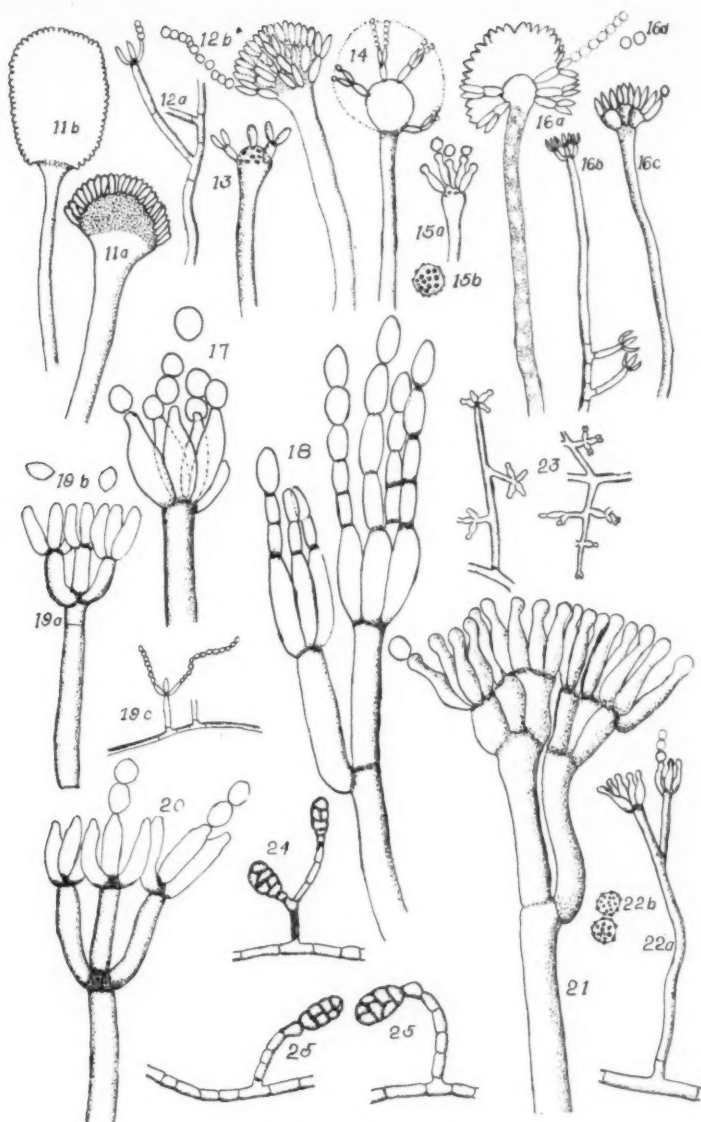
14. *Aspergillus niger*: diagrammatic sketch of conidial head, vesicle, sterigmata, and spores,  $\times 700$ .

15. *Aspergillus conicus*: a, vesicle showing the tuft-like appearance of the sterigmata,  $\times 700$ ; b, spore,  $\times 1400$ .



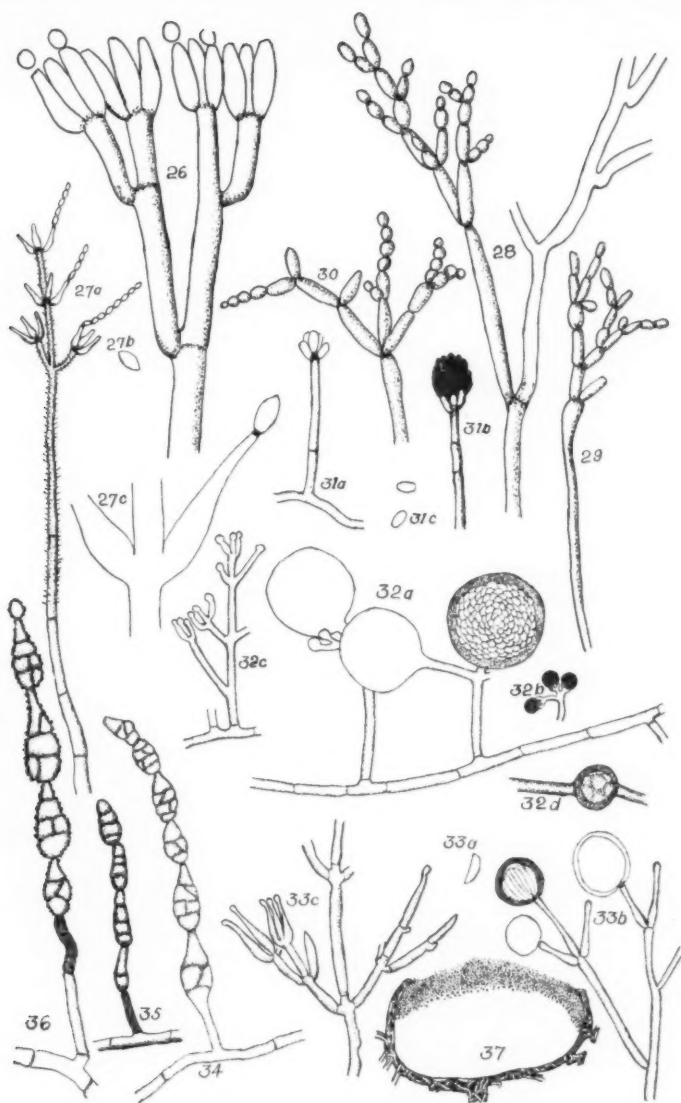
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16. *Aspergillus Sydowi*: *a*, normal vesicle, conidial head, and sterigmata (diagrammatic),  $\times 700$ ; *b*, lateral branches bearing few sterigmata without a vesicle,  $\times 450$ ; *c*, conidial head showing multiple secondary sterigmata,  $\times 700$ ; *d*, conidia,  $\times 700$ .  
17. *Aspergillus olivaceus*: conidial head,  $\times 1400$ .  
18. *Penicillium decumbens*: conidial head,  $\times 1400$ .  
19. *Penicillium roseum*: *a*, conidial head,  $\times 1400$ ; *b*, conidia,  $\times 1400$ ; *c*, lateral branching,  $\times 450$ .  
20. *Penicillium bifforme*: conidial head,  $\times 1400$ .  
21. *Penicillium atramentosum*: conidial head,  $\times 1400$ .  
22. *Penicillium echinatum*: *a*, conidiophore,  $\times 700$ ; *b*, conidia,  $\times 1400$ .  
23. *Trichoderma lignorum*: branches of the conidiophore,  $\times 450$ .  
24. *Alternaria* (No. 2): conidiophores and attached conidia,  $\times 450$ .  
25. *Alternaria* (No. 5): conidiophores and conidia,  $\times 450$ .

## PLATE 26

26. *Penicillium chrysogenum*: conidial head,  $\times 1400$ .  
27. *Spicaria elegans*: *a*, conidiophore and sterigmata,  $\times 450$ ; *b*, conidia,  $\times 900$ ; *c*, whorled arrangement of the sterigmata as lateral branches on the conidiophore,  $\times 1400$ .  
28. *Hormodendrum cladosporioides*: branched conidiophore and conidial head,  $\times 700$ .  
29. *Hormodendrum olivaceum*: conidial head and spores,  $\times 450$ .  
30. *Hormodendrum viride*: conidial head and conidia,  $\times 700$ .  
31. *Stachybotrys cylindrospora*: *a*, conidiophore attached to the sterile creeping hyphae,  $\times 450$ ; *b*, conidiophore and conidial head,  $\times 450$ ; *c*, conidia,  $\times 450$ .  
32. *Gliobotrys albo-viridis*: *a*, conidiophores as lateral branches bearing mature masses of spores,  $\times 450$ ; *b*, diagrammatic sketch of a whorl of terminal branches and the slimy mass of conidia,  $\times 140$ ; *c*, secondary branching and immature fruiting bodies,  $\times 450$ ; *d*, chlamydospore,  $\times 700$ .  
33. *Acrostalagmus albus*: *a*, conidia,  $\times 450$ ; *b*, verticillate conidiophore bearing mature conidia,  $\times 450$ ; *c*, club-shaped sterigmata,  $\times 700$ .  
34. *Alternaria* (No. 1): conidial chain,  $\times 450$ .  
35. *Alternaria* (No. 3): conidial chain,  $\times 450$ .  
36. *Alternaria* (No. 4): conidial chain,  $\times 450$ .  
37. *Coniothyrium* (species not determined): diagrammatic sketch of a cross section of the pycnidium.

(Magnifications based on original drawings which have been reduced one-half.)

## NOTES ON SOME RUSTS OF COLOMBIA

F. D. KERN AND C. E. CHARDON

Our knowledge of the rust flora of Colombia is based chiefly on the work of Dr. Eugène Mayor, who in 1913 published an excellent paper entitled "Contribution à l'étude des Uredinees de Colombie" (Mem. Soc. Neuch. Sci. Nat. 5: 442-599). In that important contribution 158 species were reported of which 84 were described as new. According to Mayor only six species had been reported from Colombia prior to 1913. His report of that year included only two of the six previously reported. Mayor's paper was based on collections made in 1910.

The following notes are based on collections made by the junior author during April, May, and June, 1926. In the data given, the collector's name and the year 1926 are omitted in order to avoid repetition since they are the same in all cases. Here are recorded 33 species. Of this number, 15 are new to the Mayor list and four, *Ravenelia Mainsiana* Arth. & Holw. on *Mimosa albida* H. & B., *Coleosporium domingense* (Berk.) Arth. on *Plumiera*, *Uredo Zeugitis* Arth. & Holw. on *Zeugites mexicana* (Kunth.) Trin., and *Puccinia abrepta* Kern on *Cyperus*, are new to South America. A fifth species, *Puccinia pallescens* Arth. on *Zea Mays* L., has been reported previously from Trinidad but this is the first record from the mainland of South America.

In the following list the 18 species which were included in the Mayor list are marked with an asterisk for ready identification. It should be pointed out that for 10 of the 18 we are not using the same names which Mayor used but full notes are included in each case so that there need be no confusion. These changes are due in some cases to transfers to different genera and in others to the combining of forms which have been provided with separate names.

In the Mayor list the species were distributed among 13 genera including the form-genera *Aecidium* and *Uredo*. In our list are only nine genera but of this number three, *Cerotelium*,

*Dicheirinia*, and *Tranzschelia*, are new to the Mayor list. *Cionothrix* is a new name but the species we are referring to this genus appeared under *Cronartium*. We have three species to include under *Uredo* but none under *Aecidium*. Our list also includes one species, *Puccinia levis* (Sacc. & Bizz.) Magn., which was known from Colombia prior to the report of Mayor and which was not rereported by him.

We are indebted to Doctor H. A. Gleason, of the New York Botanical Garden, Doctor P. C. Standley, of the U. S. National Museum, Professor A. S. Hitchcock and Mrs. Agnes Chase, of the U. S. Department of Agriculture, for aid in the determination of hosts.

1. \**COLEOSPORIUM ELEPHANTOPODIS* (Schw.) Thüm. Myc. Univ. 953. 1878.

On *Elephantopus mollis* H.B.K., near Titiribi, Dept. Antioquia, May 14, 33.

2. *COLEOSPORIUM DOMINGENSE* (Berk.) Arth. Am. Jour. Bot. 5: 329. 1918.

*Coleosporium Plumierae* Pat. Bull. Soc. Myc. Fr. 18: 178. 1902.

On *Plumiera* sp., Independence Park, Medellin, Dept. Antioquia, April 21, 13.

This species is known from a number of the West India Islands and from Panama and Guatemala. This is apparently the first report from South America. The specific name here used is founded on *Uredo domingense* Berk. Ann. Mag. Nat. Hist. II. 9: 200. 1852.

3. \**CEROTELIUM DESMIUM* (Berk. & Br.) Arth. N. Am. Fl. 7: 698. 1925.

On *Gossypium barbadense* L., Magdalena River, Santa Cruz, Dept. Bolivar, June 19, 170.

*Gossypium peruvianum* Cav., Independence Park, Medellin, Dept. Antioquia, April 21, 15.

This is a common rust known in various parts of the world on species of cotton. It was reported by Mayor under the name *Uredo Gossypii* Lagerh. It has been referred also to the genus *Kuehneola*.

4. CEROTELIUM FICI (Cast.) Arth. Bull. Torrey Club **44**: 509. 1917.

On *Ficus Carica* L., Km. 21, Tranvia de Oriente, Dept. Antioquia, May 18, 57.

This rust was originally described as a *Uredo* and has since been referred both to *Kuehneola* and *Physopella*.

5. \*CIONOTHRIX PRAELONGA (Wint.) Arth. N. Am. Fl. **7**: 124. 1907.

On *Eupatorium* sp., Magdalena River, near Barranca, Dept. Santander, June 17, 140.

This species appears in the Mayor list as *Cronartium praelongum* Wint. (Hedwigia **26**: 24. 1887). It is known from Guatemala, Costa Rica, Panama, and Veracruz on various species of *Eupatorium*, and from Brazil on an undetermined composite.

6. \*RAVENELIA INGAE (P. Henn.) Arth. N. Am. Fl. **7**: 132. 1907.

On *Inga edulis* Mart., near Venecia, Dept. Antioquia, May 26, 110.

*Inga* sp., near R. R. Station at Piedecuesta, Dept. Antioquia, May 25, 90.

This is an interesting macrocyclic species with pycnia, uredinoid aecia (primary uredo), uredinia, and telia. The spores of the uredinoid aecia are rather striking in having longitudinal striations. This stage was named twice before its telial connection was known, once by Mayor as *Uromyces porcencis* (l.c. p. 459) and again by Arthur as *Ravenelia Whetzelii* (Mycologia **9**: 64. 1917). It is this stage which is present in our material.

7. RAVENELIA MAINSIANA Arth. & Holw. Am. Jour. Bot. **5**: 426. 1918.

On *Mimosa albida* H.B.K., Fair Grounds, Medellin, Dept. Antioquia, April 18, 4.

Heretofore this species has been known only from the type locality in Guatemala. This species of *Mimosa* was not in the Mayor list.

8. *DICHEIRINIA BINATA* (Berk.) Arth. N. Am. Fl. 7: 147. 1907.

On *Erythrina glauca* Willd., Fair Grounds, Medellin, Dept. Antioquia, April 18, 6.

The uredinial stage of this species, which is the one more often collected, was named *Uredo Cabreriana* by Kern and Kellerman in 1907 (Jour. Myc. 13: 25) without recognizing its true generic connection. It is known from several localities in Central America and the West Indies and has been reported previously from South America. It is one of the commonest rusts in the vicinity of Medellin.

9. *TRANZSCHELIA PUNCTATA* (Pers.) Arth. Résult. Sci. Congr. Bot. Vienne 340. 1906.

*Puccinia Pruni-spinosae* Pers. Syn. Fung. 226. 1801.

On *Amygdalus Persica* L., near Country Club, Medellin, Dept. Antioquia, May 8, 28.

10. *\*UROMYCES APPENDICULATUS* (Pers.) Fries, Summa Veg. Scand. 514. 1849.

On *Phaseolus lunatus* L., Independence Park, Medellin, Dept. Antioquia, May 21, 81.

*Phaseolus vulgaris* L., Medellin, April 24, 21.

This species is included by Mayor (p. 462) but *Vigna luteola* is the only host mentioned.

11. *\*UROMYCES BIDENTICOLA* (P. Henn.) Arth. Mycologia 9: 71. 1917.

On *Bidens pilosa* L., Km. 21, Tranvia de Oriente, Dept. Antioquia, May 18, 61; near Fredonia, Dept. Antioquia, May 26, 104.

This species was reported by Mayor on this host and also on *Bidens bipinnata* L. and *B. squarrosa* H.B.K. but he called it *Uromyces Bidentis* Lagerh. Subsequent investigations have shown that *U. Bidentis* is a microcyclic species while *U. bidenticola* possesses uredinia. All of the Colombian specimens have uredinia and are properly referred to *U. bidenticola*.

12. \**UROMYCES COLUMBIANUS* Mayor, Mem. Soc. Neuch. Sci. Nat. 5: 467. 1913.  
On *Melanthera aspera* (Jacq.) L. C. Rich., Candela Road from Cauca Valley to "La Suiza," Dept. Antioquia, May 28, 1920.
13. *UROMYCES LEPTODERMUS* Syd.; Syd. & Butler, Ann. Myc. 4: 430. 1906.  
On *Panicum barbinode* Trin., Fair Grounds, Medellin, Dept. Antioquia, April 18, 5.  
According to Arthur (Proc. Am. Phil. Soc. 44: 207, 1925) this species is known from South America by two collections from Peru, one by Rose in 1914 and one by Holway in 1920.
14. \**UROMYCES MEGALOSPERMUS* Speg. Fungi Argent. 218. 1899.  
On *Tessaria integrifolia* R. & P., Fair Grounds, Medellin, Dept. Antioquia, April 18, 3.
15. \**UROMYCES PROËMINENS* (DC.) Pass. Rab. Fungi Eur. 1795. 1873.  
On *Chamaesyce hirta* (L.) Millsp., Medellin, Dept. Antioquia, April 24, 22; Hacienda Marsella, Cauca Valley, Dept. Antioquia, May 27, 116.  
This species was included by Mayor under the name *Uromyces euphorbiicola* (Berk. & Curt.) Tranzschel (Ann. Myc. 8: 8, 1910) which we regard as a synonym. He did not include this host.
16. *PUCCINIA ABREPTA* Kern, Mycologia 11: 140. 1919.  
On *Cyperus* sp. (possibly *C. caracasanus* Kunth.), Hacienda Marsella, Cauca Valley, Dept. Antioquia, May 27, 115.  
This appears to be the first report of this species from South America. Our specimen agrees well with the type, which is from Costa Rica, especially in having the pores of the urediniospores covered with a swollen hyaline cuticle.
17. \**PUCCINIA CENCHRI* Diet. & Holw.; Holw. Bot. Gaz. 24: 28. 1897.  
On *Cenchrus echinatus* L., Magdalena River, Santa Cruz, Dept. Bolivar, June 19, 169.

This species has been reported also from Brazil (see Arth. Proc. Am. Phil. Soc. 44: 158, 1925).

18. \*PUCCINIA CRASSIPES Berk. & Curt. Grevillea 3: 54. 1874.

On *Ipomoea* sp., Candela Road from Cauca Valley to "La Suiza," Dept. Antioquia, May 28, 1921.

Only aecia are present in this specimen. The same is true of the specimen reported by Mayor (l.c. p. 488) which he calls *Puccinia Ipomoeae-panduratae* (Schw.) Syd. Arthur has shown that the Schweinitz name *Aecidium Ipomoeae-panduranae* was founded on an *Albugo*. The proper name for the *Ipomoea* rust seems to be as above.

19. PUCCINIA EVADENS Harkn. Résult. Sci. Congr. Bot. Vienne 343. 1906.

On *Baccharis cassinaefolia* DC., Km. 21, Tranvia de Oriente, Dept. Antioquia, May 18, 1921.

In the Mayor list there are seven different species of rust reported on as many species of the genus *Baccharis*. It is with hesitation that we report still another, for *Puccinia evadens* Harkn. is not in this list, although it has been reported from South America. On the other hand ours is a different species of host and the characters agree so well with *Puccinia evadens* that the present reference seems entirely justifiable.

20. PUCCINIA GRAMINIS Pers. Neues Mag. Bot. 1: 119. 1794.

On *Agrostis perennans* (Walt.) Tuckerm., Km. 21, Tranvia de Oriente, Dept. Antioquia, May 18, 1921.

Arthur reports (Proc. Am. Phil. Soc. 44: 178, 1925) twelve hosts from South America for this common grass rust. He comments that this is not a long list for a rust that is so common in the north temperate zone. *Agrostis perennans* is not in the Arthur list.

21. \*PUCCINIA HETEROSPORA Berk. & Curt.; Berk. Jour. Linn. Soc. 10: 356. 1869.

On *Sida spinosa* L., Medellin, Dept. Antioquia, April 24, 1921.

22. PUCCINIA LEVIS (Sacc. & Bizz.) Magn. Ber. Deuts. Bot. Ges. 9: 190. 1891.

On *Axonopus scoparius* (Fl.) Hitchc., Finca Mirasol, near Sabaletas, Dept. Antioquia, May 20, 72.

*Paspalum pilosum* Lam., Finca Mirasol, near Sabaletas, Dept. Antioquia, May 20, 75.

A fairly common rust in southern United States, Central America, the West Indies, and northern South America.

23. \*PUCCINIA MELAMPODII Diet. & Holw.; Holw. Bot. Gaz. 24: 32. 1897.

*Puccinia Synedrellae* P. Henn. Hedwigia 37: 277. 1898.

*Puccinia Eleutherantherae* Diet. Ann. Myc. 7: 354. 1909.

*Puccinia Wedeliae* Mayor, Mem. Soc. Neuch. Sci. Nat. 5: 528. 1913.

On *Synedrella nodiflora* (L.) Gaertn., Fredonia, Dept. Antioquia, May 25, 96.

*Wedelia caracasana* DC., Medellin, Dept. Antioquia, May 7, 27.

It is our opinion that the microcyclic rusts on *Eleutheranthera*, *Synedrella*, and *Wedelia* are the same species and that they are referred properly as here indicated.

24. PUCCINIA PALLESCENS Arth. Bull. Torrey Club 46: 111. 1919.

On *Zea Mays* L., near Venecia, Dept. Antioquia, May 26, 109.

This species has pale yellow uredinia and nearly colorless walls in the urediniospores as compared with the cinnamon-brown uredinia and golden- or cinnamon-brown walls of the common corn rust, *Puccinia Sorghi*. Arthur has reported *Puccinia pallescens* from Trinidad (Am. Phil. Soc. 44: 156) but this appears to be the first report from the mainland of South America.

25. \*PUCCINIA POLYGONI-AMPHIBII Pers. Syn. Fung. 227. 1801.

On *Persicaria punctata* (Ell.) Small (*Polygonum acre* H.B.K.), near Medellin, Dept. Antioquia, April 24, 19; near Titiribi, Dept. Antioquia, May 14, 32.



26. PUCCINIA PSIDII Wint. Hedwigia **23**: 171. 1884.

On *Jambos Jambos* (L.) Millsp. (*Eugenia Jambos* L.), Independence Park, Medellin, Dept. Antioquia, April 21, 18; Poblado Sorrento, Dept. Antioquia, May 16, 45.

27. \*PUCCINIA ROTUNDATA Diet. Hedwigia **36**: 32. 1897.

On *Vernonia patens* H.B.K., Candela Road from Cauca Valley to "La Suiza," Dept. Antioquia, May 28, 123.

This rust was reported by Mayor (l.c. p. 511) on the same host, under the name *Puccinia rugosa* Speg. (Anal. Soc. Ci. Argent. **17**: 92, 1884). Although this is an older name than *Puccinia rotundata* it is not valid as there is a *Puccinia rugosa* Billings, 1871, which is another thing.

28. PUCCINIA RUELLIAE (Berk. & Br.) Lagerh. Tromsø Mus. Aarsh. **17**: 71. 1895.

On *Blechnum Blechnum* (L.) Millsp., Road from Medellin to Itagui, Dept. Antioquia, April 18, 8.

A common rust in tropical regions of both hemispheres.

29. \*PUCCINIA SOLANITA (Schw.) Arth. Mycologia **14**: 19. 1922.  
*Puccinia solanicola* Mayor, Mem. Soc. Neuch. Sci. Nat. **5**: 505. 1913.

On *Solanum* sp., Itagui, Dept. Antioquia, April 18, 10.

The Schweinitz specific name *solanita* was founded on a specimen from Surinam and dates back to 1853. We believe that Mayor's *Puccinia solanicola* is the same and must become a synonym as indicated.

30. \*PUCCINIA SORGHI Schw. Trans. Am. Phil. Soc. II. **4**: 295. 1832.

*Puccinia Maydis* Bereng. Atti Sci. Ital. **6**: 475. 1845 (hyponym).

On *Zea Mays* L., Itagui, Dept. Antioquia, April 18, 9; Km. 21, Tranvia de Oriente, Dept. Antioquia, May 18, 60.

Apparently a common rust wherever Indian corn is grown.

31. UREDO CHERIMOLIAE Lagerh. Bull. Soc. Myc. Fr. **11**: 215. 1895.

On *Annona Cherimolia* Mill., Medellin Park, Dept. Antioquia, April 16, 2; near Titiribi, Dept. Antioquia, May 28, 126.

This species is known from Ecuador and Florida on this host, and from Cuba, Florida, and Yucatan on other species of the genus *Annona*.

32. \*UREDIO CUPHEAE P. Henn. Hedwigia 34: 99. 1895.

On *Parsonsia Pinto* (Vand.) Heller (*Cuphea Balsamona* C. & S.), near Titiribi, Dept. Antioquia, May 14, 31; Km. 21, Tranvia de Oriente, Dept. Antioquia, May 18, 69.

This species is known also from the West Indies and Brazil.

33. UREDIO ZEUGITIS Arth. & Holw.; Arth. Am. Jour. Bot. 5: 538. 1918.

On *Zeugites mexicana* (Kunth.) Trin. (*Senites mexicana* Hitchc.), Km. 21, Tranvia de Oriente, Dept. Antioquia, May 18, 52.

This appears to be the first report of this species for South America and also the first report for this species of host. It is known from Guatemala on *Zeugites Hartwegi* Fourn.

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## ON THE OCCURRENCE OF BIFLAGELLATE SWARM CELLS IN CERTAIN MYXOMYCETES

FRANK A. GILBERT

(WITH 2 TEXT FIGURES)

While the swarm cells of Myxomycetes have almost invariably been described as uniflagellate, occasionally swimmers with two flagella have been reported, associated with normal swarm cells. These biflagellate forms are of two types, the first being characterized by having two flagella at the anterior end and the second by having one flagellum at each end. Swarm cells of the first type, as far as the writer has been able to ascertain, have been recorded only by DeBary<sup>1</sup> who observed them in a culture of the spores of *Trichia varia*. Swarm cells of the second type however, are of more frequent occurrence, Constantineau<sup>2</sup> having found them in cultures of the spores of *Dictydium cancellatum*, Durand,<sup>3</sup> of *Enteridium Rozeanum*, and DeBary,<sup>1</sup> of *Fuligo septica* and *Trichia varia*, in which, as above indicated, they were associated with swarm cells of the first type.

While studying spore-germination<sup>4</sup> in many representative genera and species of Myxomycetes, the writer also has observed biflagellate swarm cells. Both types occurred in his cultures, which were made by sowing the myxomycetous spores in distilled water in Syracuse glasses.

Swarm cells of the first type, that is with two flagella at the

<sup>1</sup> Bary, H. A. De. Die Mycetozen. Zeitschr. Wissensch. Zoologie 10: (1860).

<sup>2</sup> Constantineau, L. Ueber Entwicklungsbedingungen der Myxomyceten. Ann. Myc. 4: 495 (1906).

<sup>3</sup> Durand, E. J. Notes on the Germination of *Enteridium Rozeanum*. Bot. Gaz. 19: (1894).

<sup>4</sup> The writer is greatly indebted to Dr. William H. Weston, Jr., for his help during the course of the work and also for invaluable aid in the preparation of the manuscript.

anterior end, were found in cultures of one gathering of *Stemonitis fusca* from the Cambridge region. About twenty-five per cent of the swarm cells from this gathering had two flagella while the remaining seventy-five per cent were normal. The second flagellum was not apparent in living specimens except under the highest powers of the microscope (TEXT FIG. 1, e-f), but could easily be seen when the organisms were killed and stained. A number of Van Tieghem cell cultures were made and in each was placed a single spore taken from a sporangium of the previously mentioned gathering. In a few of these cultures, the swarm cell that emerged had two flagella at one end, indicating that this type is truly myxomycetous and, since other gatherings of *Stemonitis fusca* developed only normal swarm cells, must be considered as merely an abnormal form.

Swarm cells of the second type, that with a flagellum at each end, were found a number of times by the writer. They occurred sporadically in cultures of various genera and species of Myxomycetes, but were not present in cultures of *Enteridium Rozeanum* in which species they had previously been reported by Durand. Because of the irregularity of their occurrence, it seemed of interest to determine definitely their nature and origin. Two explanations seemed possible, first, that they were truly myxomycetous, either aberrant swarm cells of the species sown or normal swarm cells of some contaminating species, and second, that they were Protozoa which in some manner had made their way into the cultures. Protozoa of various sorts are not infrequently found in myxomycete cultures, since their cysts, like the spores of bacteria and fungi, may be blown about and come to rest on sporangia; or according to Pinoy<sup>5</sup> may even be carried up by the rising plasmodium, and incorporated with them during their formation. As a result, these cysts, as well as various other foreign bodies, may eventually be introduced in the cultures with the spores of the Myxomycete that is being studied.

As far as the general characters are concerned, swarm cells of this type seem to correspond very closely to normal swarm cells

<sup>5</sup> Pinoy, E. Rôle des Bactéries dans le Développement de certains Myxomycètes. Ann. Inst. Pasteur 21: 622 (1907).

in size, color and general shape (TEXT FIG. 1, g-i). The nucleus, however, which in normal swarm cells of the Myxomycetes is usually apparent at the anterior end below the base of the flagellum (TEXT FIG. 1, a-c), appears, in these biflagellate forms,

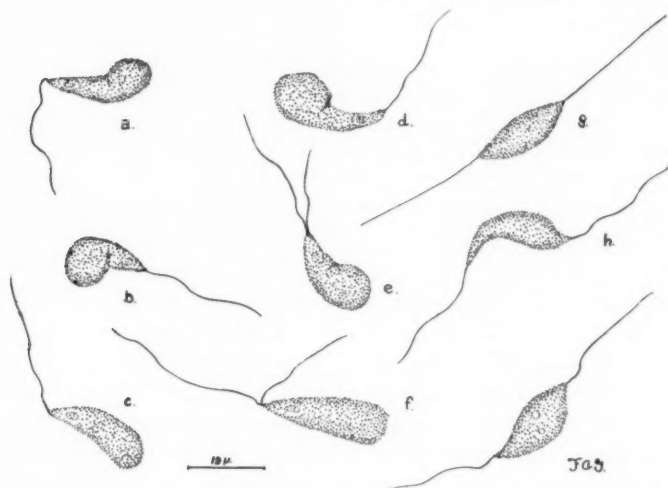


FIG. 1. a. *Hemitrichia clavata*. Swarm cell, showing its form during the rotating movement which is simulated by the protozoan, *Cercomonas*; b, c. *Trichia affinis*. b. Swarm cell, showing form during rotating movement. c. Swarm cell, showing form during creeping movement which is not simulated by *Cercomonas*; d-f. *Stemonitis fusca*. d. Normal swarm cell. e, f. Swarm cells from the same gathering as d, with two flagella at the anterior end; g-i. *Cercomonas longicauda*. g. Protozoan, showing form during the smooth gliding movement. h, i. Protozoan, showing form during the rotating movements.

to be nearer the center of the cell and is distinguished with great difficulty. Their movements, owing to the characteristic rotation which they frequently exhibit, simulate very closely the similar phenomenon so frequently observable in myxomycetous swarm cells generally. The latter, however, are easily differentiated by the fact that they frequently pass to a creeping or vermicular stage (TEXT FIG. 1, c), which has never been observed in biflagellate "swarm cells" of the type under consideration. The biflagellate forms, on the other hand, are clearly distinguished

from the normal myxomycetous swarm cells by a characteristic motion peculiar to themselves. They often glide smoothly forward in an absolutely straight line, with the flagella stretched out from each end (TEXT FIG. 1, g), a movement which, as far as has been observed, never occurs among myxomycetous swarm cells. These differences in methods of progression clearly differentiate the two organisms when they are examined under the higher powers of the microscope.

In order to study them further, a number of the biflagellate individuals of this type were separated from cultures of *Hemitrichia clavata*, in which they had appeared, placed in sterile watch glasses in water, a little autoclaved hay added, and the whole set aside for a few days. At the end of this time, in the original cultures, the normal uniflagellate *Hemitrichia* swarm cells, which are very susceptible to unfavorable environmental conditions, had become microcysts or had changed to myxamoebae; while the forms with a flagellum at each end, in all of the original cultures in which they occurred, as well as in the autoclaved hay cultures separated from them, in contrast to the myxomycetous swarm cells associated with them, had not modified their characteristics in any respect. Indeed, in the course of a number of experiments, they were found to form cysts only under conditions, for example, of extreme heat or dryness.

The most striking difference between the two forms, however, is in the method of division. All myxomycetous swarm cells which were observed in the many cultures of the writer, divided in the normal manner<sup>6</sup> by retracting the flagellum and rounding off, before the separation in two parts (TEXT FIG. 2, a-c). The biflagellate forms, on the contrary, accomplished their division without any such retraction, one of the persistent flagella being eventually attached to each daughter cell (TEXT FIG. 2, d-i). The process by which this division takes place is as follows: the original cell slowly elongates and a slight constriction appears

<sup>6</sup>F. X. Skupienski in his "Recherches sur le cycle évolutif de certains Myxomycètes," Paris (1922), mentions a division of the swarm cells of *Didymium nigripes* by longitudinal fission which is anomalous among the Myxomycetes.

at the middle (TEXT FIG. 2, *e*); the constriction gradually becomes more marked until the cell at this point is about one fourth of its original diameter (TEXT FIG. 2, *f*); the two halves then gradually draw apart and as they do so remain connected

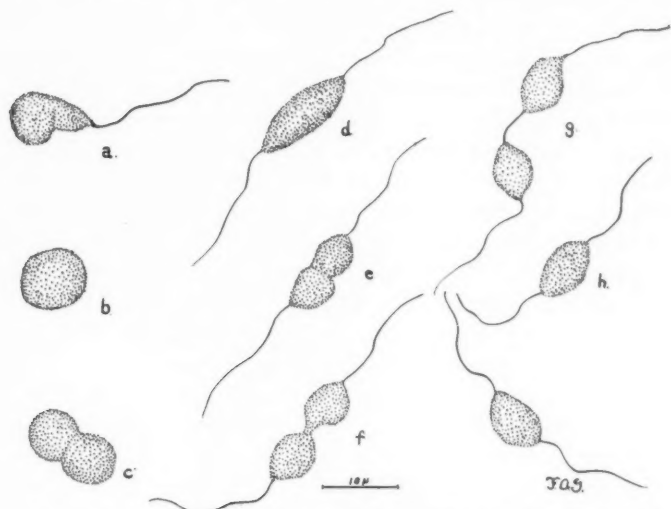


FIG. 2. *a-c. Hemitrachia clavata*. *a*. Swarm cell. *b*. Swarm cell just before division, with the flagellum absorbed. *c*. Swarm cell during division; *d-i. Cercomonas longicauda*. *d*. Protozoan before division. *e-i*. Stages in division, showing the flagella, which are not absorbed but remain active during the process.

by a slender protoplasmic thread (TEXT FIG. 2, *g*), which soon breaks (TEXT FIG. 2, *h, i*), and each daughter cell, supplied with a new flagellum, swims away in its characteristic biflagellate condition. This division is of the type known to occur among the monad flagellates in certain genera such as *Cercomonas*<sup>7</sup> and also in the Labyrinthulales, an order generally regarded as belonging to the more primitive Mycetozoa. This order was investigated by Zopf<sup>8</sup> who found that "die Amöbae sich

<sup>7</sup> Dallinger & Drysdale. Researches on the Life History of a Cercomonad. Mon. Mic. Jour. 10: (1873).

<sup>8</sup> Zopf, W. Zur Kenntniss der Labyrinthuleen, einer Familie der Mycetozoen. Beiträge zur Physiologie und Morphologie tieferer Organismen. Leipzig (1873).

ziemlich bedeutend streckt und dann etwa in der Mitte einschnürt, worauf beide Theile auseinander rücken, aber durch einen Hyaloplasmastrang verbunden bleiben."<sup>9</sup> Zopf also found that the flagella of the swarm cells of that order fuse with one another or with the body of an adjacent swarm cell almost immediately after germination<sup>8</sup> to form a net plasmodium.

It would seem, therefore, that the second of the two possible explanations of their presence, which has already been suggested above, was likely to be the true one, namely that they should be regarded as protozoan contaminants, accidentally introduced. That they cannot be referred to any member of the Labyrinthulales seems indicated by their failure to show the slightest tendency to form a net plasmodium. On comparing their characters with those of the various well-known types of Protozoa, they appear to correspond in their structure, behavior, and methods of reproduction to *Cercomonas longicauda* Duj.,<sup>10</sup> a monad which seems to be common in moist vegetable materials, like decaying wood. For a verification of this determination, the writer is indebted to Dr. J. A. Dawson of the Zoological Department of Harvard University.

Since neither of the three authors above cited, who have reported the occurrence of bipolar swarm cells, records any observations on their division, it cannot be definitely assumed that they were dealing with structures identical with those studied by the writer. In view, however, of their frequent

<sup>9</sup> This similarity of division presumably indicates a close relationship between the Monads and the Labyrinthulales. In the latter group, however, the cells partly fuse into what Zopf calls a "Fadenplasmodium" and eventually form sori of spores, thus throwing their true affinity with the higher Myxomycetes, between the Monads and which they possibly form a connecting link. Some authors, however, are of the opinion that the resemblance between the net plasmodium of the Labyrinthulales and the true plasmodium of the Myxogastrales is entirely superficial and shows no relationship whatsoever between the two orders.

<sup>10</sup> Fr. Stein in "Der Organismus der Infusionsthier," 3: Leipzig (1878), shows longitudinal division in this species and transverse division in others of the same genus. The organism studied by the writer, however, agreed so closely with the description and measurements of *Cercomonas longicauda* Duj. that it can only be identified as such even though its division, conforming to that typical for the genus, does not agree with the anomalous method recorded by Stein.



appearance in numerous cultures of five or more different species, it seems highly probable that they may have been identical with the type above described. However this may be, it seems rather clearly indicated that these bodies have, in reality, no direct connection with the Myxomycetes, but are merely contaminations of accidental origin. This assumption is borne out by the fact that no such bodies have ever been seen actually to escape from a germinating spore, and further that they have occurred in cultures, for example, of decaying wood in which the presence of myxomycetous spores was at least not apparent.

It seems logical to conclude therefore, from the above investigations, that former reports of myxomycetous swarm cells with a flagellum at each end may have been based on the confusion of these Protozoa with normal swimmers.

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LOPHODERMIIUM INFECTANS MAYR A  
SYNONYM OF HYPODERMA  
ROBUSTUM TUBEUF

J. S. BOYCE

(WITH 1 TEXT FIGURE)

When Mayr, in the latter part of the last century, made a trip through the forests of North America he collected or observed a number of parasites which he later named in his book (1) on these travels, but without describing the organisms, so now it is difficult and in some cases impossible to determine just what he has recorded, since the only clue to the identity of the parasite is the host on which it occurs.

Among his collections was a needle-inhabiting fungus parasitic on white fir (*Abies concolor* (Gord.) Parry) in the San Bernardino Mountains of California. The perithecia and spores which appeared on needles from 2 to 6 years old were not mature, according to Mayr's statement, in this collection made in November 1887. He named the fungus *Lophodermium infectans* n. sp. without describing it. Tubeuf (2, p. 16) who later studied some of Mayr's collections did not find this parasite among them, but did find a fungus belonging to the same family, Hysteriaceae, on a species of fir, but the material was without label. Tubeuf decided the host was *Abies concolor* and described the parasite as a new species, *Hypoderma robustum*. The genus *Hypoderma* was formerly included in *Lophodermium*.

Among duplicate collections from the Farlow Herbarium of Harvard University received by the writer through the courtesy of Professor Roland Thaxter was one labelled "*Lophodermium infectans* Mayr on *Abies concolor*. Type of Mayr. See his Waldungen." As far as can be ascertained this material came to Dr. Farlow directly from Mayr. A study of the collection shows the fungus to be the same as Tubeuf named *Hypoderma robustum*. The measurements of the spores without the gelatinous sheath as given by Tubeuf were  $3 \times 30\text{--}36 \mu$ , while in the

collection from the Farlow Herbarium 50 spores ranged from  $3-7 \times 18-34 \mu$  with an average of  $4 \times 29 \mu$ , not a significant

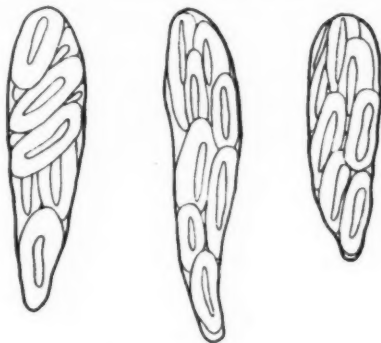


FIG. 1. Asci and spores from the collection of *Lophodermium infectan* received from the Farlow Herbarium.

difference. Asci and spores from this collection are shown in figure 1.

From the foregoing it seems highly probable that the collection studied and described by Tubeuf was originally labelled *Lophodermium infectans* by Mayr but the label was later lost. The only doubt is Mayr's statement that the fungus was not mature when he observed it, but it is not unusual when examining these fungi to find both mature and immature perithecia in the same collection. Since Mayr's name is a nomen nudum, *Hypoderma robustum* Tubeuf stands.

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- (1) **Mayr, H.** Die Waldungen von Nordamerika. xii + 448 pp., 16 figs., 12 pls., Munich. 1890.
- (2) **Tubeuf, Carl von.** Studien über die Schüttekrankheit der Kiefer. In Arbeiten aus der Biologischen Abtheilung für Land- und Forstwirtschaft am Kaiserlichen Gesundheitsamte, v. 2, no. 1, pp. i-ii + 1-160, figs. 1-32, pls. 1-7. 1901.

## MISCELLANEOUS COLLECTIONS OF NORTH AMERICAN RUSTS

WILLIS R. HUNT

Listed below are fifty specimens of rusts, representing nine genera and thirty species. These were collected by the writer on various travels (not collecting trips) outside of the New England states during the past two years. Specimens of all of these collections have been deposited in the Connecticut Agricultural Experiment Station Herbarium.

In the spring and fall of 1925 the writer spent a few days in the Pocono Mountains, Matamoras, Pike County, Pennsylvania. Of the twenty-three collections made, the following rusts on the specific hosts are not listed in North American Flora:

*Coleosporium Solidaginis* on *Solidago neglecta*, *S. rugosa* and *S. serotina*, *Puccinia Andropogi* on *Chelone glabra*, *P. Grossulariae* on *Ribes Cynosbati*, *P. Urticae* on *Carex crinata* var. *gynandra*, *P. Violae* on *Viola conspersa*, *Uromyces hybridi* on *Trifolium hybridum*, *U. Lespedezae-procumbentis* on *Lespedeza capitata*.

In the fall of 1925 while doing some collecting in Maine, a trip was made over the line into Canada. *Puccinia graminis* on *Phleum pratense* was collected in St. Come. It is not reported in North American Flora from Quebec on this host.

A trip to Bermuda was made in the winter of 1926. In as far as the writer knows, *Uromyces Fabae* on *Vicia Faba*, and *Coleosporium Solidaginis* on *Solidago sempervirens* have never been reported from these islands. The rust on windsor bean was collected in the garden of the Agricultural Experiment Station by courtesy of Dr. Lawrence Ogilvie, Government Plant Pathologist.

The collections made at Ithaca while attending the International Congress of Plant Sciences have all been previously reported from New York by Arthur.

CAEOMA NITENS Schw. On *Rubus* sp.: I, Matamoras, Penna., May 30, '25.

COLEOSPORIUM SOLIDAGINIS (Schw.) Thüm. On *Aster cordifolius* L.: II, Enfield Falls, N. Y., Aug. 17, '26. On *Solidago arguta* Ait.: II, Taugkannock Falls, N. Y., Aug. 20, '26; II, Enfield Falls, N. Y., Aug. 17, '26. On *S. neglecta* T. & G.: II-III, Matamoras, Penna., Sept. 17, '25. On *S. rugosa* Mill.: II, Matamoras, Penna., Sept. 17, '25. On *S. sempervirens* L.: II, Bermuda, March 3, '26. On *Solidago serotina* Ait.: II, Matamoras, Penna., Sept. 17, '25.

COLEOSPORIUM DELICATULUM (Arth. & Kern) Hedge. & Long. On *Solidago graminifolia* (L.) Salisb.: III, Matamoras, Penna., Sept. 18, '25.

GYMNOCONIA INTERSTITIALIS (Schl.) Lagerh. On *Rubus* sp.: I, Matamoras, Penna., May 30, '25.

GYMNOSPORANGIUM BERMUDIANUM (Farl.) Earle. On *Juniperus bermudiana* L.: III, Bermuda, March 3, '26.

KUEHNEOLA ALBIDA Magn. On *Rubus* sp.: II-III, Matamoras, Penna., Sept. 17, '25.

MELAMPSORA AMERICANA Arth. On *Salix* sp.: II-III, Matamoras, Penna., Sept. 18, '25; II, Ithaca, N. Y., Aug. 17, '26.

MELAMPSORA MEDUSAE Thüm. On *Populus tremuloides* Michx.: II-III, Matamoras, Penna., Sept. 18, '25.

PHRAGMIDIUM POTENTILLAE-CANADENSIS Diet. On *Potentilla canadensis* L.: III, Matamoras, Penna., Sept. 17, '25.

PUCCINIA ANDROPOGI Schw. On *Chelone glabra* L.: 0-I, Matamoras, Penna., May 30, '25.

PUCCINIA BARDANAE Corda. On *Arctium minus* Bernh.: II-III, Matamoras, Penna., May 30, '25; Sept. 18, '25; II, Ithaca, N. Y., Aug. 18, '26.

PUCCINIA CLEMATIDIS (DC.) Lagerh. On *Agropyron repens* (L.) Beauv.: II, Victor, N. Y., Aug. 21, '26.

PUCCINIA GRAMINIS Pers. On *Phleum pratense* L.: II, Matamoras, Penna., Sept. 17, '25; II, St. Come, Quebec, Sept. 25, '25; II, Victor, N. Y., Aug. 21, '26.

PUCCINIA GROSSULARIAE (Schum.) Lagerh. On *Ribes Cynosbati* L.: 0, Matamoras, Penna., May 31, '25.

PUCCINIA HELIANTHI Schw. On *Helianthus annuus* L.: II-III, Paterson, N. J., Sept. 20, '25.

PUCCINIA HIERACII (Schum.) Mart. On *Cichorium Intybus* L.: II, Middle Hope, N. Y., July 25, '25; II-III, Bermuda,

March 4, '26. On *Taraxacum officinale* Weber: II, Matamoras, Penna., May 31, '25; II, Ithaca, N. Y., Aug. 18, '26.

PUCCINIA LANTANAE Farl. On *Lantana involucrata* L.: III, Bermuda, March 4, '26.

PUCCINIA MALVACEARUM Bert. On *Althea rosea* Cav.: III, Westown, N. Y., June 1, '25; III, Middle Hope, N. Y., July 25, '25; III, Ithaca, N. Y., Aug. 18, '26.

PUCCINIA MENTHAE Pers. On *Monarda fistulosa* L.: II-III, Matamoras, Penna., Sept. 18, '25.

PUCCINIA SUAVEOLENS (Pers.) Rostr. On *Cirsium arvense* (L.) Scop.: II, Ithaca, N. Y., Aug. 18, '26.

PUCCINIA THALICTRI Chev. On *Thalictrum polygamum* Muhl.: III, Enfield Falls, N. Y., Aug. 17, '26.

PUCCINIA URTICAE (Schum.) Lagerh. On *Carex crinata* var. *gynandra* (Schwein.) Schwein. & Torr.: II-III, Matamoras, Penna., Sept. 18, '25.

PUCCINIA VIOLAE (Schum.) DC. On *Viola conspersa* Reichenb.: 0-I, Matamoras, Penna., May 30, '25; *Viola cuculata* Ait.: II, Tarrytown, N. Y., July 11, '24.

UROMYCES FABAE (Pers.) DeBary. On *Vicia Faba* L.: II, Bermuda, March 4, '26.

UROMYCES HYBRIDI Davis. On *Trifolium hybridum* L.: III, Matamoras, Penna., May 30, '25; II, Middle Hope, N. Y., June 25, '25.

UROMYCES JUNCII-EFFUSI Sydow. On *Juncus effusus* L.: III, Matamoras, Penna., June 1, '25.

UROMYCES LESPEDEZAE-PROCUMBENTIS (Schw.) Curt. On *Lespedeza capitata* Michx.: III, Matamoras, Penna., Sept. 17, '25.

UROMYCES MEDICAGINIS Pass. On *Medicago lupulina* L.: II, Bermuda, March 3, '26.

UROMYCES TRIFOLII (Hedw. f.) Lév. On *Trifolium pratense* L.: II, Ithaca, N. Y., Aug. 18, '26; II, Victor, N. Y., Aug. 21, '26.

UROMYCES TRIFOLII-REPENTIS (Cast.) Liro. On *Trifolium repens* L.: I, Matamoras, Penna., May 30, '25; II, Marlboro, N. Y., July 10, '24.

CONN. AGR. EXP. STATION

## WHY THE DIFFERENCES IN PUBLISHED SPORE-SIZES?

C. H. KAUFFMAN

In a recent paper (Bull. Soc. Myc. Fr. 42: 43-50. 1926) by my good friend and honored colleague Dr. René Maire of France, that experienced and thorough mycologist has reviewed the causes that underlie the often confusing and divergent spore-measurements given by different authors for the same species. He discusses these causes under four categories:

1. Errors of determination.
2. Changes in the spore due to drying or to reagents.
3. Faults due to the technique used in measuring.
4. Innate variations of the spores themselves.

The detailed discussion of these categories is so well done and covers so many common and important cases that I am anxious that all American workers with fungi, mycologists and phytopathologists alike, should have their attention called to this valuable paper. There is scarcely one of us who has not been guilty, either unconsciously or from ignorance or in moments of haste, of neglecting one or more of the precautions which Maire here sets down.

Passing over the first category, an example of which is the mis-determination of a plant and therefore recording its spore measurements under a species name to which it does not belong, I shall briefly discuss some of the others. Two considerations are involved when one measures the spores of a herbarium specimen, *i.e.* a dried-out plant. First, the procedure used in reestablishing the normal turgidity of the spores, and, secondly, giving the reader of the paper in which such spore-measurements are printed a statement of the reagent used and the manner of its use. In all of my own published work, a fourth or a half of a one per cent solution of potassium or sodium hydroxide was invariably used with dried specimens. Unfortunately, I have scarcely ever called attention to this fact in my papers. In all

of my papers dealing with agarics since the appearance of the "Agaricaceae of Michigan," all spore-measurements of my own recorded therein were made from the fresh plant when collected, except when type specimens are mentioned, in which case the procedure involved the use of the hydroxide. Maire very properly emphasizes the importance of noting the type of reagent used.

Under his third category, he discusses: (a) the inclusion of immature spores in obtaining the range in size; (b) mistaking the central large globule of many spores for the whole spore, when its wall is very thin and transparent; (c) the use of micrometer values which are not exact; (d) errors due to the use of a defective micrometer scale. Experienced students will not generally "fall down" on the first two counts, and yet it is surprising how often spore-measurements are sent in to me with specimens, where the discrepancies which appear in our measurements are nearly always due to this lack of discrimination between young and mature spores. Maire gives an account of a very prominent French mycologist, E. Boudier, who had the unusual experience of having used a defective micrometer scale for a while. The proper calibration of one's micrometer is such a self-evident matter that it may be passed over here.

One could easily write an extensive paper on the variability of spores. Dr. Maire's remarks are well worth most careful consideration by all who are concerned in the question of spore-size. Although his discussion is limited to the Basidiomycetes, there is much that applies to spores of other groups of fungi.

The stimulus which caused Maire to review the question of spore-size came from a paper by C. E. Martin in *Bulletin de la Société Mycologique de Genève*, No. 9, 1925. The latter author is quoted as strongly advising that when a mycologist critically studies an older named plant with the microscope he should write the name and its author in the usual way, but attach in addition his own name or that of the modern author whose microscopical data agree with his, in order that there may be no question of the proper determination, at least as to microscopic points. Maire says he has used this idea for a long time in his notes. It was also a feature of the fine work of Ricken. In



my own publications, I have also for a long time both in my notes and in print employed this means of fastening my identified plant to the species as known to a particular modern author, instead of broadly writing only Schaeffer, or Bulliard, Persoon or Fries after it; for, in the latter case, my microscopic data would not be vouched for and must stand alone. These more careful modern methods, now in use by the most experienced mycologists, will do more in my judgment to stabilize the species of the Basidiomycetes than any amount of legislating.

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## NOTES AND BRIEF ARTICLES

Since publishing our article on *Poronia leporina*, Dr. Roland Thaxter, of Harvard, has communicated to us two additional specimens. One was collected at Cocoanut Grove, Florida, in 1897 and the other from Sandy Run, South Carolina, in 1903. This adds something to our knowledge of the distribution of this interesting fungus and it is hoped that these notes will stimulate mycologists to make a further search for this species.

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A number of models of fleshy fungi have recently been installed in the museum of the New York Botanical Garden by Dr. Fred J. Seaver. These models were made by Dr. Joshua Rosett, the process being a new one which he himself has devised. It is impossible to display fleshy fungi satisfactorily either by means of dried specimens or those preserved in liquid. There are many different types of models each of which has its own special advantage. This is the first attempt of our museum to install models of living plants and it is hoped that this collection may be added to and improved upon until we have a fairly complete display of these evanescent forms of plant life, commonly known as the fungi.

### ERWIN F. SMITH—FRIEND OF YOUTH

It seems desirable to relate briefly an incident concerning Dr. Erwin F. Smith, who recently passed away. In 1916, shortly after the publication of my first paper (dealing with an insect gall), I happened to meet him for the first time. He spent considerable time showing me some of his crown gall slides, telling me about his work, and discussing my "maiden effort." Picture to yourself the impression made on an unknown youngster receiving such attention from an internationally famous scientist. Having become interested in a scheme I had to produce galls artificially, he set aside a part of his already overcrowded laboratory for my use and installed a sink with water connections.

This he did, although I was employed in an entirely different government bureau having no connection with the United States Department of Agriculture. (A record of this work will be found in the *Zeitschrift für Pflanzenkrankheiten*, v. 34, pp. 344-346, 1924.)

It is a pleasure to record this episode, because I have heard it said that Dr. Smith was self-centered. May the science of Plant Pathology become richer and richer in such self-centered individuals. H. R. ROSEN.

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As this issue goes to press, we have received notice of the death of Professor Bruce Fink, who was found dead in his laboratory on the morning of July 10. America thus loses one of her best known lichenologists. Dr. Fink at the time of his death was professor of botany in Miami University, Oxford, Ohio. He has been an associate editor of *MYCOLOGIA* from its beginning. A more extended article on his life and work will doubtless appear in a later issue.

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News has also just been received of the death of Dr. Lars Romell of Stockholm, Sweden, on the night of July 12. Dr. Romell is well known to mycologists the world over and has served as an associate editor of *MYCOLOGIA* from its beginning. A more detailed account will appear in a later issue.

